Risk mitigation for Mediterranean fruit flies with special emphasis on risk reduction for commercial imports of clementines (Several varieties of *Citrus reticulata*) from Spain

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I. EXECUTIVE SUMMARY

This document examines risk mitigation measures to prevent the introduction of the Mediterranean fruit fly in imports of citrus from Spain. We used a quantitative (probabilistic) simulation approach to evaluate how offshore measures coupled with cold treatments assured reduced risks compared to the use of quarantine cold treatments only. The overall system includes the pathway (importation of citrus) that may result in pest introduction. The procedures proposed by Plant Protection and Quarantine (PPQ) ensure a systematic examination of the hazards and the identification of critical phases (i.e., application of key phytosanitary practices) of the overall system that are key to mitigating risks. We analyzed the production system and characteristics of the pest; we then evaluated the different mitigation practices; and identified critical control points. This document describes how the proposed additional mitigation practices reduce introduction risks. We concluded that two elements (critical control points) are fundamental to the successful reduction of risks associated with the importation of citrus fruit from Spain: the limitation of the population of pests in the field and the application of quarantine cold treatments such that probit 9 mortality is approximated. Probit 9 refers to treatments that result in ca. 99.9968 percent mortality. This corresponds to a survival rate of 0.000032 (0.0032 percent) of all individuals exposed to a treatment that is said to achieve Probit 9 mortality. In addition to critical control points, supplementary phytosanitary measures (e.g., surveys, port inspections, quality assurance, training, field trapping, and management of the pest in other hosts; US domestic fruit fly trapping, and others) provide additional safeguards that result in risk reductions that further diminish the potential effects of uncertainties and variability inherent in the system. Even in the unlikely event that all containers were to encounter suitable conditions (hosts, susceptible fruit, climate), the probability of a mated pair arriving anywhere in the United States in the course of a year was low. Our analysis showed that the likelihood of a mated pair in fruit from Spain was less than one in two thousand years, considering the 95th percentile of the distribution (less than one in more than ten thousand years using the mean of the distribution), even assuming multiple containers shipped to suitable areas. The probability of a mated pair in a single container was less than one in a million (95th percentile of distribution). This document concluded that proposed new mitigation practices (notably, assuring low field populations of fruit flies) reduced overall risk compared to the current system of cold treatments alone.

II. INTRODUCTION

This document evaluated the risk mitigation measures associated with the commercial importation of Spanish clementines. The approach was to first identify system components, evaluate pest attrition associated with each component, and to examine the effects of the combination of components (field practices, post-harvest treatments, and other safeguarding practices) in minimizing the likelihood of pest introduction. We used a stochastic approach to analyze the percentage of infested fruit and associated fruit fly larvae as it is managed with the procedures represented within each component of the pathway. A stochastic approach allows inclusion of variability associated with a system. Variability is included by consideration of the range of possible values, in addition to mean or most likely value estimates for responses associated with each system component. The endpoint of this analysis included estimation of the probability that a pair of fruit flies occurred in a given container. Throughout this text, a "container" is defined as consisting of a 40-foot conveyance, usually carried individually by truck and used in the transport of clementines fruit.

To emphasize components of the system that are of greatest safeguarding value and subject to control, we identified those components (critical control points) that upon failure would likely imperil the safeguarding objectives. Thus, we identified critical control points, which we detail. The identification of these critical control points permits focus on those components that are key to the overall safeguards.

Citrus from Spain has been exported to the United States for some twenty years, beginning in the early 1980s (Snell, personal communication). Events during 2001 (finding live, apparently viable larvae in Spanish clementines) led to the suspension of the rule that allowed shipments to occur. Enhanced mitigations were outlined in an updated work plan, the key elements of which are described and analyzed here. A workplan is a detailed description of requirements and mandatory phytosanitary practices that must be met before commercial movement is allowed to proceed. Key elements of the work-plan include activities that are key to the successful safeguarding against pests associated with this pathway. The work plan (D. West, personal communication) limits populations of pests in the groves in Spain in addition to requiring the application of cold treatment as modified and updated by technical reviews during February 2002 (http://www.aphis.usda.gov/oa/clementine/coldtxre.pdf). The present risk mitigation document was motivated by revisions required to existing rules which must be based on a reassessment of the mitigations associated with this pathway. This document is part of the regulatory process; it evaluated whether the mitigations proposed resulted in reduced risks compared to the existing (baseline) system.

III. REGULATORY BACKGROUND

The permit for the importation of clementines from Spain was based on evaluations of risk associated with this commodity some twenty years ago (Imai, personal communication). Regulatory authority for importation of citrus from Spain is described in 7 CFR 319.56. However, recent evidence suggests that the reliance on a single tactic, cold treatment T107, USDA 1998, which was assumed to provide quarantine security may not allow for some variances in the application of the tactic. This observation is based on the occurrence of apparently viable, live fruit fly larvae detected in cold treated Spanish clementines in 2001. The precise cause of the finds during 2001 are not known, but they may include an atypically warm year and very early season in Spain that allowed for early population build-up coupled with late season warm temperatures that exacerbated the problem. Evidence of higher than normal populations during 2001 compared to other years was provided by the trap capture information for 2000 and 2001 (MAPA 2001), which indicated higher trap catches in 2001 than in 2000, and by the multiple finds of apparently viable, live larvae in fruit from Spain during 2001 (Snell, personal communication). The higher than average temperatures, which occurred in 2001compared to 2000 (MAPA 2001), are also consistent with a high population during 2001. As noted before, in addition to high field populations, variability in quarantine cold treatment may have also been a factor in the occurrence of live larvae after treatment. Variability in management procedures in different groves constitutes yet another potential risk element. The identified risk elements are being addressed with new and updated mitigation practices reflected in the new workplan. The key feature of the new workplan is the mandatory limitation of field densities below a specified threshold. The other key component of the workplan, cold treatment, already existed in the previous workplan.

The approach of USDA to address risk mitigation is multi-pronged: manage potential variation in the application of cold treatment and other phytosanitary phases with increased quality control at all stages, but with particular emphasis on critical control points, addressing field population levels through a series of pre-harvest mitigation practices and fruit cutting with rejection of lots if live larvae are found (for details, see our published proposed rule). The revision of cold treatment schedules has already resulted in an update to the treatment recommendations, specifically by lengthening the duration of the treatment (http://www.aphis.usda.gov/oa/clementine/coldtxre.pdf).

We chose a systematic approach to implement verifiable risk mitigation measures consistent with our risk analysis guidelines (e.g., USDA 2000), which describes the assessment of risk. The PPQ guidelines are consistent with the International Plant Protection Convention's International Standard for Phytosanitary Measures (ISPM 11, "Pest Risk Analysis for Quarantine Pests"; http://www.fao.org/ag/agp/agpp/PQ/Default.htm). Other risk analysis guidelines exist that emphasize risk management and the identification of critical control points (that is, identification of those components of the system that must be carefully controlled to minimize risk).

To emphasize components of the system that are of greatest risk mitigation value and subject to control, we refer to some concepts from the area of food safety. Specifically we reviewed concepts associated with a risk management approach developed by NASA, FDA, the Pillsbury Company and others called "Hazard Analysis and Critical Control Point" (HACCP) (Buchanan 1990, Corlett, 1991, Guzewich 1987, ICMSF 1989, NACMCF 1992, Pierson and Corlett, 1992).

For the present system, we identified critical control points in HACCP as being equivalent to critical control points in the area of phytosanitary safety and as identified in the workplan. Critical control points are components of the safeguarding system that assure that risks are minimized and which are subject to subsequent verification by PPQ. The identification of these control points permits focus on those components that are key to the overall safeguards. The adoption of HACCP concepts provides valuable framework and guidelines, while not constituting a departure from existing procedures. A HACCP plan largely parallels "work plans" developed as part of regulatory procedures that allow commodities to move internationally. We emphasize that the reference to HACCP does not represent a departure from existing guidelines but rather an extension or refinement that reflects more emphasis on key management aspects (e.g., critical control points). In contrast to commodity risk assessments as described in the PPQ guidelines (USDA, 2000), the present document emphasizes the evaluation of risk management approaches. A more detailed description of how HACCP principles can be applied to phytosanitary risk management is shown in appendix 1.

IV. DESCRIPTION OF MEDITERRANEAN FRUIT FLY BEHAVIOR AND DYNAMICS

The biological description of *Ceratitis capitata,* the Mediterranean fruit fly or Medfly, presented here is not meant to be an exhaustive review, but emphasizes characteristics of the pest that are key to understanding risk mitigation. The Medfly is a pest of fleshy fruit, which occurs in tropical and subtropical areas, and is one of the most destructive fruit pests in the world, due to its broad host range, and its ability to survive and expand its range.

Medfly infests more than 250 types of fruits, flowers, vegetables, and nuts. Weems (1981) lists 42 host species as "heavily or generally infested," 15 species as "occasionally infested", 25 species as "rarely infested", 21 species as "laboratory infestations", and 153 species as "unknown importance". Liquido *et al.* (1991) report 180 genera, worldwide, as hosts for this insect.

Female Medfly oviposit up to 14 eggs below the skin of the host fruit (McDonald and McInnis, 1985), with the potential of producing up to 1000 eggs throughout its lifetime. Hatching occurs in 2-18 days, (depending upon the temperature), the three larval instars require 6-50 days, pupation occurs in soil, with adult eclosion in 6-60 days (EPPO, 1979; Weems, 1981).

Adults fly short distances but may be carried by wind for 2.4 km or more (PNKTO 18, 26; Weems, 1981). Steiner *et al.* (1962) have reported migratory movements of 40-72 km, and sustained over-water flights of 19-64 km. This insect is multivoltine, with 10-15 generations possible in warm climates (EPPO, 1979). Bodenheimer (1951) has recorded the following developmental ranges for various stadia: at 20^{o}C: egg 9.7 days, larvae 53.6 days, pupa 79.1 days; 35^{o}C: egg 1.0 days; larva 4.7 days; pupa 7.2 days; developmental zero occurred at 10.5^{o}C, 9,8^{o}C and 9.7^{o}C, respectively, for egg, larva and pupa. Adult flies cannot live more than one to two weeks below 5^{o}C.

In Spain, the Medfly has been known since the XIX century. Management procedures are necessary in most production areas and most years to reduce populations (Azcarate-Luxan 1996). Up to eight generations may occur in Spain and damage may be great if left unmanaged (Agusti 2000) but four to six are more common (Planes and Carrero, 1996). Management practices include the use of population monitoring, mass-trapping, bait sprays, biological control and broadcast sprays (aerial and terrestrial) (Agusti 2000, Dominguez 1998).

V. CITRUS PRODUCTION IN SPAIN

Citrus production has been an important element of the Spanish economic sector since its introduction in the VII century; indeed, by the 1500s, references to citrus production are common. There are records of large commercial citrus production from the area of Mallorca by the end of the nineteenth century that cite 30,000 ha of citrus, a large proportion of which were destined for exports mostly within Europe (Azcarate-Luxan 1996). Mandarins were introduced into Valencia from Italy in 1845 (Agusti 2000). Clementines varieties of mandarins have been known only since the 1950s in Spain with most of the recent varieties originating in the 1960s and 1970s (Agusti 2000).

Citrus is produced in different provinces bordering the Mediterranean Sea as shown in Figure 1. Approximately 271,000 ha are in production for both the domestic and export markets (MAPA 2001).

Production regions for citrus (in descending amount of surface area dedicated to citrus, in hectares) include: Valencia 183,000; Andalucia 45,000; Murcia 33,000; Cataluna 6,300; Baleares Islands 2,300; Canary Islands 1,300 (MAPA 2001).

Citrus production in Spain continues to focus on a large export market. Spain exports large quantities of citrus to most of Western and Eastern Europe; Russia, Argentina, Australia, Brazil, Canada, United States, and Iceland. Exports from Spain to the United States date from 1985 (W. Snell and D. West, personal communication). Exports from the 2001 season were interrupted by reports of live, apparently viable larvae in fruit mostly in US distribution outlets but also in confirmation samples by PPQ personnel (D. West, personal communication and first-hand observer of samples with live larvae). Exports worldwide were 1,248,515 tons in 1994 and 1,121,162 tons in 1996. Total mandarin (including clementines) exports to the United States totaled 12,848 tons in 1994; 15,172 tons in 1995, 23,107 tons in 1996 (MAPA 1999). During the 1999-2000 season, exports to the United States were approximately 80,000 tons (source: Appendix 2).

VI. DESCRIPTION OF MEDFLY POPULATION DYNAMIC IN SPAIN

The exact date of introduction of the Medfly into Spain is unknown; however there are records of this pest from the nineteenth century (Azcarate-Luxan 1996), although its introduction likely pre-dates that period. The Medfly is reportedly common along the Mediterranean coast (Dominguez 1998). Dominguez (1998) stated that reports of Medfly from the interior (i.e., away from coastal areas) are largely due to the movement of produce from coastal areas. He notes that there are no damage reports from Castilla La Vieja and that the colder regions in the Central and inland portions of Spain are probably not suitable for the continuous presence of the Medfly.

Agusti (2000) reported up to eight generations possible depending on the weather. Planes and Carrero (1996) reported from four to six as more common. Hosts in Spain include peaches, apricots, pears, persimmons ("caquis"), oranges, and mandarins (Agusti 2000, Planes and Carrero 1996, Dominguez 1998). Peach is reported the preferred host in Spain (Dominguez 1998). Clementines are not optimal hosts; the maximum survival rates of Medfly in citrus under optimal temperature conditions are ca. 9% in late oranges and ca. 8% in clementines (Santaballa et al. 1999).

Agusti (2000) summarized the dynamics of the Medfly in Spain noting that a first generation may occur during the winter, developing in late season oranges and mandarins, especially in more protected sun-warmed areas. In spring, a subsequent generation (second) attacks apricots and peaches. The third generation appears at the beginning of summer in peaches. Two more generations (fourth and fifth) may develop during August and September on peaches, pears, figs, and persimmons at the same time that it may also attack the earliest varieties of oranges and mandarins. During the fall, another generation (sixth) develops on oranges and mandarins. Additional generations are possible if fall and winter temperatures are warm. Dominguez (1998) presents a similar description of host phenology. Agusti (2000) reported that Medfly does not attack citrus before September-October under field conditions. The reason is first, because other preferred hosts are present; and second, because the condition of citrus at this time (color and hardness) are not adequate for Medfly oviposition. Dominguez (1998) notes that the colder months are most likely spent in the soil in the pupal stage.

VII. REVIEW OF CONTROL PRACTICES IN SPAIN

<u>Field controls</u>. Control practices reported in Spain against the Medfly in the nineteenth century included collecting fallen fruit and burying it after covering with lye ("cal viva") (Azcarate-Luxan 1996). More recently, integrated pest management methods have included the use of classical biological control, mass trapping, pesticide bait sprays, population monitoring, and others (Agusti 2000, Planes and Carrero

1995). At this time the main control tactics include the use of bait sprays triggered by a threshold amount of flies caught in "Nadel" traps baited with an attractant, usually Trimedlure©. The threshold that triggers bait sprays is 0.5 flies/trap/day (Planes and Carrero 1995). The use of large numbers of traps as a mechanism of control (without bait sprays) is also cited. With relatively high densities of traps, the percentage of fruit that is infested is 0 to 20% (Planes and Carrero 1995). Planes and Carrero (1995) cite the placement of traps in preferred hosts and before citrus begins maturation (e.g. in apricots and peaches in April) to allow for management of the pest population such that subsequent population build-up and economic damage are avoided.

Culling and Packing house Controls. Culling occurs during many phases beginning at harvest when blemished fruit is usually removed. Direct inspection and culling for quality control then occurs during at least two other phases (quality control and packing) (APHIS site visit report 2001).

Cold Treatment. The use of cold temperatures to destroy fruit flies has long been the subject of research (e.g., Back and Pemberton 1916; Yothers and Mason 1930; Petty and Griffiths, 1931; Mason and McBride 1934; Nel 1936, 1937). More recent research has refined and expanded the use of cold treatment to many more species and with a variety of equipment and conditions that all result in mortality close to 100% (e.g., Sproul 1976; Hill et al., 1988; Santaballa et al. 1999). Nearly one century of experience in the movement of different commodities from infested to non-infested areas attest to the effectiveness of cold treatments; however, the experiences during 2001 with Spanish clementines (i.e., the occurrence of live larvae in US markets after cold treatment) suggest that variability in the application of this control may be responsible for less than Probit 9 mortality if careful quality control procedures are not followed. Probit 9 refers to treatments that result in ca. 99.9968 percent mortality. This corresponds to a survival rate of 0.000032 (0.0032 percent) of all individuals exposed to a treatment. Prior to 2001, port inspections after cold treatment suggested that densities of dead larvae were below 1% in most inspected cargo (W. Thomas, personal communication). Updates to the cold treatment schedules required by USDA (http://www.aphis.usda.gov/oa/clementine/coldtxre.pdf) and a series of quality control procedures and inspections are expected to account for variability within the system (Gould *et al*., 2002).

Regulatory and phytosanitary practices in Spain. The use of national regulatory programs has a long history in Spain. There exist detailed reports of national control programs dating back to the XVI century focusing on locust management (Azcarate-Luxan 1996).

The current (2002) Spanish production and agricultural regulatory system has some elements that are similar to the American system of state autonomy and federal coordination of some export programs. Specifically, the Spanish Ministry of Agriculture, Fisheries, and Nutrition (MAPA) has coordination responsibilities for phytosanitary issues, especially related to export systems and the management of invasive species. However, the implementation of recommendations and domestic programs lies with regional administrative units called "Comunidades Autonomas". These autonomous communities (AC) roughly correspond to the divisions presented in figure 1.

As part of its responsibilities and according to specific regulations associated with citrus management, the ACs directly monitor some 800 Medfly traps in Valencia (ca. 1 trap per 200 Ha) distributed in the citrus production regions. Additional traps are managed at the farm level. MAPA also issues recommendations regarding field controls, packinghouse quality control and cold treatments. Actual field activities are monitored and carried out by the administration of the ACs and exporters. In addition to field controls (especially aerial sprays), individual farmers may also use supplementary traps and ground pesticide applications to manage localized problems. Close contacts with academia assure the application of technologically advanced research activities (Artolachipi, Cortina and Esteruelas, Santaballa personal communication). Other management practices are described in Appendix 2.

VIII. RISK REDUCTION MEASURES AGAINST FRUIT FLIES

Spanish clementines have been imported into the United States under CFR 7-319.56. However, the events of 2001 (live, apparently viable larvae reported in imported fruit) have motivated the establishment of additional safeguards and the addition of several new quality control activities to the program. Key elements of the revised work-plan are detailed below.

Key phytosanitary measures

- Traps will be used to monitor adult populations and placed in preferred hosts.
- A preharvest field certification/management plan will be implemented to control the field Medfly populations to reduce the infestation rate of fruit to below detectable levels of 1½ % after harvest.

- Fruit cutting at the inspection site and prior to cold treatment (in Spain) will include the cutting of sufficient fruit to allow detection of densities 1½ % or greater of infested fruit). This step is intended to allow a maximum 1½ % level of infestation (percent infested fruit) with a confidence level of 95% (Steel and Torrie, 1980). This is considered a critical control point.
- All fruit will be traceable to its source or production unit throughout the entire system.
- Cold treatment as per schedule T107 (USDA, 1998) will be implemented, as revised in http://www.aphis.usda.gov/oa/clementine/coldtxre.pdf.
- Upon arrival in the United States, fruit will be inspected according to the guidelines provided in the USDA inspection guidelines (USDA 1993) and applicable updates.

The above is a partial listing of the safeguards considered as part of the workplan. The most important components of the system from a phytosanitary perspective are those that permit monitoring of critical steps ("control points") in the pathway. A low level of the pest in the field and application of cold treatment are the most important phytosanitary measures for this pathway.

In addition to the work plan safeguards offshore; domestic safeguards (USA) include increased awareness at ports of entry and a review of national trapping protocols. To assure quality control over the long term, harvest crews, quality control personnel and others involved in this safeguarding system will be trained in the identification of fruit fly punctures and other Medfly evidence. Training is already part of PPQ's New Officer Training program. This document does not contemplate beginning a new training system since one already is in place; it does however, emphasize the need for periodic retraining and updates.

The work-plan contains numerous changes intended to increase the overall effectiveness of the treatment and to provide for more stringent quality assurance and quality control. However, the key component of the new mitigations is the limit placed on field densities. This is justified at least two ways: first, the empirical evidence reported by PPQ in 2001 (finding numerous apparently viable and live larvae in clementines from Spain) suggested that field densities during the 2001 season were high; second, the mortality due to cold treatment as expressed here induces a proportional decrease in the potential survivors. This implies that the absolute number of live larvae that survive treatment is density-dependent. That is, the more larvae initially (that is the higher the infestation rate), the more total larvae that are likely to survive after treatment.

Quality Control. Quality control procedures will be integrated into standard operating procedures. Quality control will be co-developed by USDA and MAPA and managed by the ACs for the on-site QA procedures related to trapping and survey in Spain; Packinghouse culling and sampling; and other field activities. Quality control procedures will be managed and monitored by PPQ and include stringent review of cold treatment and port of entry inspections. All procedures will be part of a work-plan that will be dynamic and subject to review, especially in response to new evidence and new techniques. The purpose of a dynamic work-plan is to ensure the maintenance of safeguards equivalent to or greater than implemented initially.

IX. ASSESSMENT OF RISK REDUCTION AND EVALUATION OF OVERALL SYSTEM RISKS

Figure 2 outlines the main components of the system. Circles in figure 2 describe the main control points. Circles with bold lines identify critical control points. The first circle in the system represents all fruit destined for export. The next circle represents field pest population (fruit flies) after different management practices (e.g., bait sprays, fallen fruit removal, ground spot treatments, mass trapping, and others) have been implemented. This circle ("Infested fruit in field") constitutes a critical control point. The next circle accounts for the fact that there may be multiple fruit flies developing within a fruit. This circle includes all factors that limit the potential number and viability of these pests due to host and environmental interactions. The next circle ("Flies after cold treatment") refers to the flies in fruit after in-transit cold treatment; it is the second critical control point. The next circle identifies port inspections at US ports of entry as another filtering mechanism where fruit will be inspected and rejected if live larvae are found. Whereas they are a mitigation component, parameters for inspection at port of entry were not available at the time of this writing. The next circle includes the effect of dilution away from suitable areas. That is, not all citrus that is imported ends up in states where susceptible hosts occur or where conditions for establishment prevail. Medfly is not likely to become established in an area where citrus does not grow. Areas that have winter temperature too cold for citrus are also too cold for the pest. Citrus is generally the only good host available in subtropical or Mediterranean climates during the late winter or early spring (Miller 1992). The endpoint ("Probability of a mated pair in a container") is directly related to the likelihood that fruit flies become introduced (established and spread).

We used probabilistic simulation (Vose 2000; Olkin et al. 1994) to concentrate on the behavior of the critical components included in figure 2. The export program was evaluated and the results are detailed below. Two systems are contrasted, the baseline system that represents the production and export system as it existed in 2001 and the new, proposed mitigation program. The new mitigation program includes limitation on the proportion of infested fruit prior to cold treatment, increased quality control and efficacy of the cold treatment. The present characterization of the two systems resulted in identical parameters for both systems with the only difference being in the component that describes fruit infested in the field. That is, we assume that the main quantifiable differences between the baseline and the new program are the limitation of field densities under the new program. Other differences are assessed qualitatively.

Component 1 (C1). Number of Fruit Shipped (number of fruit per container and total amounts per year)

This component evaluates the amount of fruit that is exported in a single a container (a "shipment" in previous versions). We also review here the amount of total fruit that is exported in one year to the United States from Spain.

There may be 20-25 clementines per 2.5 kg carton. Each pallet of fruits contains 360 cartons, and each 40 ft. container holds 20 to 21 pallets (USDA, 1987; W. Thomas, personal communication; Santaballa, personal communication). If we assume that the number of fruit per carton follows a uniform distribution U (20,25), the sum of the 7,380 cartons (360x20.5) per container would follow a normal distribution (by Central Limit Theorem, N (166,050;15,375). The upper 95% confidence interval for the number of clementines in one container would be less than 166,294.

Exports from Spain to the United States date from 1985 (W. Snell and D. West, personal communication). Export totals worldwide were ca. 1,248,515 tons in 1994 and 1,121,162 tons in 1996 (MAPA, 1999). Total mandarin (including clementines) exports to the United States totaled 12,848 tons in 1994; 15,172 tons in 1995, 23,107 tons in 1996 (MAPA 1999). During the 1999-2000 seasons, exports to the United States were ca. 80,000 tons (Appendix 2).

As noted above and in terms of total fruit shipped per year, there were ca. 80,000 tons shipped during 2000. We had based previous analyses on that observation. However, a larger number was used in a separate economic analysis: 116,406 metric tons. We increased our estimate of the maximum number of yearly containers to match. Thus, a maximum 6408 individual 40 ft containers of clementines per year was considered in this version of the analysis.

Most fruit is transported domestically as containerized cargo, typically traveling from the port of entry to distribution center(s) and finally to retail outlets. Throughout this analysis, we assume that one container is the basic biologically relevant unit for which risk must be evaluated. This is consistent with research reports (e.g., Landolt et al. 1984).

Landolt *et al*. (1984) proposed shipments (commercial containers) as a logical unit for which risk should be assessed. They stated: "The most practical point to assess the risk of an introduction occurring is the probability of a potential mating pair or gravid female…getting through quarantine. A potential mated pair might be defined as a nonsterile male and a nonsterile female occurring in the same area during the same period such that mating is possible. For our purposes, a pair of fruit flies emerging from the same shipment would be considered a potential mated pair. The additional problems of survival, feeding, dispersal, mate finding and host finding are unknown but add a large degree of safety beyond the probability of a mated pair occurring. The risk of an introduction should then be calculated as the probability of one or more mated pairs per shipment surviving quarantine measures". Although a conservative estimate, it is also possible to estimate the probability of mated pairs assuming that multiple containers end up in suitable locations assuming hosts and environments are always suitable. We included multiple container effects using the approach of Whyte *et al*. (1996) and Wearing *et al*. (2001).

We assessed the probability of mated pairs in an individual container and for all containers in one year (Vail *et al*., 1993) as detailed below. The estimation of the probability of at least one mated pair in multiple containers ($P_{multiple}$) was estimated as $P_{multiple} = 1-(1-P)^S$, where P is the probability of a mated pair in one container and S is the number of all containers (Whyte *et al*., 1996; Wearing *et al*. 2001). We assumed that there were 166,050 fruit per container (e.g., a 40 ft container, see USDA 1987).

Fruit flies are relatively poor fliers (e.g., Weems, 1981), with maximum known flights of up to 64 km (ca. 40 miles according to Steiner et al., 1962) and commonly reported to move lesser distances (Weems, 1981, PNKTO 18,26). Most fruit containers will arrive in unsuitable areas such that flies that may emerge will not be able to find suitable host, suitable climates, find mates and lead to successful introductions, thus the estimates of fruit arriving in the United States were further divided below (Component 5) into those that arrive in areas that are suitable vs. those that arrive in areas that are unsuitable.

Component 2 (C2). Fruit Infested with Larvae in the Field

This component represents the proportion of fruit that is infested with fruit flies in the field and that arrive in the packinghouse. We have assumed that the final proportion of infested fruit in the packinghouse (that is, the fruit that will be shipped and treated) is represented by this component.

Fruit fly management in the field results in reduced pest densities. The proposed system (see section on “Key phytosanitary measures”), further assures that field pest densities do not go beyond a threshold level, a maximum 1.5% infested fruit or equivalent measure of low density. A maximum threshold (critical control limit) is assured by a sampling system that uses fruit cutting and visual inspections at the packinghouse that result in 95% confidence that populations are less than or equal to (but no greater than) 1.5% (AQIM 2001, Steel and Torrie, 1980). The minimum expected pest infestation proportion is 0% infested fruit. Prior to 2001, port inspections did not find live larvae in citrus from Spain on commercial shipments (Thomas, personal communication).

A Pert distribution (Vose 2000) was constructed to represent our knowledge about this component. The minimum value was zero, the most likely value was 0.001 and the maximum value was 0.015 (1.5 %) for the mitigated scenario. We could have also chosen zero as minimum and most likely based on zero observations between 1985 and 2000. For the baseline, there is no limit as to field populations as part of the importation program; the maximum is then established by empirical and documentary evidence (e.g., Agusti, 2000; EPPO, 1979; Weems, 1981; Planes and Carrero, 1995; Santaballa and Moner, personal communication) that suggest that up to 15% percent of fruit may be infested. That maximum value was the only difference between the baseline and the new mitigation scenario. This maximum value (15%) appears to be associated with unmanaged or abandoned hosts.

After our initial estimates described above, we evaluated more direct, recent evidence of infested Spanish clementines from intensive sampling during 2001. The sampling results from USDA-APHIS-PPQ and cooperators, especially from the state of California are shown in Tables 1-3.

We acknowledge that dissection of fruit is not likely to find all infested fruit. For example, Gould (1995) reported that sampling for Caribbean fruit flies in grapefruit resulted in an average 35% of the infested fruit being found. This suggests that sampling for medfly in Spanish clementines is also not likely to find all infested fruit. We note however that clementines are smaller fruit than grapefruit and have therefore a much larger surface area to inspect. Clementines are also easier to dissect than grapefruit. Finally, the Spanish clementines were sampled by teams of inspectors, often from different agencies working in cooperation. Cooperative work implies that a significant amount of cross checking takes place and that a higher proportion of infected fruit is likely to be detected. Indeed, the experiences from 2001 when USDA and State inspectors successfully detected Medfly larvae even though very low densities were present, is indicative of very high efficiency and ability by agricultural officials to find larvae in infested fruit as part of routine inspections.

The year 2001 was assumed to have higher relative populations as per MAPA 2001 that indicated higher trapping densities compared to other years. The sampling from vessels in 2001 (Table 1) found a proportion of infested fruit of 0.0016 (95% upper and lower confidence limits assuming a beta distribution of 0.001 and 0.002, respectively). The sampling from distribution outlets nationwide had a lower proportion of infested fruit of 0.0005. We note however that we did not have adequate unbiased quantitative data to estimate accurate differences between 2001 and previous years. Comparisons were thus mostly indirect resulting from relative estimators.

If we assume that the reports of Gould (1995) apply to clementines, then a likely infestation rate of clementines was 0.0016 divided by 0.35 for samples from vessels inspected in 2001. That results in a proportion of 0.00457 (ca. 5 per thousand) infested fruit prior to cold treatment. Our examination of additional evidence resulted in estimates consistent with or indicative of lower infestations than our original estimates (summarized in the second paragraph of this section, C2). We did not revise our estimates downward because the samples were obtained without the benefit of statistical design, the effect of culling at packinghouse/distribution centers was unknown, and thus these estimates are empirical indicators, subject to confirmation with subsequent sampling. However, the empirical evidence does suggest that the value for this component may be an overestimate, subject to confirmation with subsequent observations. Confirmation of this value of an overestimate will have an effect on the overall calculations. That effect will be to reduce estimations of risk of introduction.

Finally, whereas the statistical inference is that a 95% confidence limit around the 1.5% value may indeed permit infestation levels higher than 1.5%, the actual likelihood of this value being greater than 1.5% was not considered realistic given additional evidence and the manner in which the workplan is managed (specifically, there are additional mitigations that a limited proportion of all lots inspected may be infested with proportions greater than 1.5%, such that finding a greater proportion than that established in the workplan will shut down the entire export program). This assumption of a maximum of 1.5% was thus based on the ease with which high populations are detected with current sampling plans, based on the empirical evidence that suggests that real infestations are much lower than 1.5%, based on USDA's site visits to Spain, and on personal communications as noted.

Component 3 (C3). Larvae per fruit

This component accounts for the fact that there may be several larvae in an infested fruit. Amounts of total medflies can then be estimated by multiplying the total number of larvae per fruit times the proportion infested fruit times the fruit that make up a container.

The survival of eggs to viable adults in citrus was reviewed by Santaballa et al. (1999) under optimal temperature conditions. This component emphasizes the number of larvae per fruit that will lead to viable adults. Even at optimal conditions, the survival of immature forms to viable adults did not reach 10%. The reports from Spain that citrus are not optimal hosts are consistent with findings reported by Leyva et al. (1991) who studied a closely related tephritid, *Anastrepha* sp. in oranges (a larger fruit than clementines) and other hosts. Leyva *et al*. (1991) reported lower suitability of citrus hosts compared to peaches. Among the citrus hosts tested, grapefruit was the most satisfactory host and Valencia oranges the least satisfactory. Leyva *et al*. (1991) also noted that although high numbers of larvae may be associated with laboratory-infested fruit, e.g., more than 97 larvae per fruit, these high infestation rates do not result in proportionally higher numbers of adults. Leyva *et al*. (1991) use the term "overinfested" to describe the relationship between high number of larvae and mortality of most larvae, pupae, and adults associated with these high infestations per fruit. Leyva *et al*. (1991) report an average three larvae per fruit with a maximum of 17 and a minimum of one in oranges. The reports on *Anastrepha* are consistent with our observations at the ports and include the observed range. McDonald and McInnis (1985) reported that Medfly might deposit up to 14 eggs below the skin of host fruit. As noted before, Santaballa et al (1999) showed that not all immature flies survive to adults. We also note that this reproductive strategy (i.e., many eggs that do not all results in viable adults) is very common among insect (Gillot, 1980).

The number of larvae per infested fruit was initially estimated from the evidence in PNKTO (18,26); Leyva *et al*., 1991; Santaballa et al. 1999; McDonald and McInnis, 1985; and W. Thomas, personal communication). In order to determine a most likely value, we reviewed interception records and interviewed port inspectors (W. Thomas, personal communication). The most common maximum number of larvae reportedly found in infested fruit was ca. 15. However, it must be noted that this was based on a very small number of observations of actual larvae in fruit. Early versions of this analysis used a most likely value of three larvae per fruit, a maximum of 17 and a minimum of one, as parameters for this component. We used a Pert distribution to simulate associated variability. No differences were assumed between the baseline and the new program scenarios. The maximum was later modified (see discussion below).

In addition to our initial estimates detailed above, we obtained direct sampling evidence from 2001 (Tables 1-3). This evidence is about total larvae in fruit and suggests that the average larvae per fruit vary from four to twelve. Evidence from Gould (1995) suggests that inspectors dissecting grapefruit found 9.5% of actual Caribbean fruit fly larvae in grapefruit. That would suggest that actual larvae densities could vary from 40 to 120 if Gould (1995) is an appropriate model. The evidence from Santaballa et al. 1999) showed that ca. 10% of all immature forms will achieve adulthood; then the number of viable larvae (those that will survive to adults) is roughly the same as the number detected by inspectors (four to twelve). This direct evidence and its interpretation suggest that the value originally proposed (three to seventeen) is consistent with or greater than that suggested by additional evidence. We acknowledge that the observed values (e.g. Table 3) may be greater than the value used as a most likely estimate (i.e., three). The direct observations (e.g. Table 3) do not reveal the viability of these larvae (i.e., the proportion that actually survives to produce a viable adult); therefore, we used the values as recorded in table 4 and supported with the evidence as noted.

Comments to an earlier draft of the study suggested that the number chosen as maximum (17 viable adults resulting from larvae) was questionable. Comments suggested that we review data specifically related to clementines. Such data was available from the Spanish experimental evidence (Santaballa, 1999). They noted a maximum actual survival of 7.65% for Medflies in clementines. Assuming that field infestations lead to 100 eggs per fruit (clearly higher than possible in larger sized fruit such as tested by Leyva et al. (1991)), this would result in a maximum eight viable adults resulting from larvae. We adjusted our estimates of the maximum to reflect this lower number (see table 4).

Component 4 (C4). Cold Treatment

This component represents the effects of the cold treatment. The fruit is treated with refrigeration that approximates probit-9 mortality (second critical control limit). The evidence from cold treatment studies shows that at most 32E-6 (32 in a million) individuals will survive. As per requirements detailed in USDA's Treatment Manual (USDA 1998), both the Medfly and species of *Anastrepha* can be controlled using a combination of different temperatures and periods that all result in the required mortality. These treatments (T107 series) required different temperatures if different periods are used. Specifically for treatments (prior to changes in the requirements in 2002) at a temperature setting of 32^{o}F or below the required exposure period was ten days; at 33^{o}F or below the period was 11 days; at 34^{o}F or below it was 12 days; at 35^{o}F or below it was 14 days; at 36^{o}F or below it was 16 days. Note however, that as part of the review of cold treatment efficacy these treatments were modified to assure increased mortality.

These temperature requirements imply the existence of critical limits, which are a related to the period of exposure. Therefore, critical limits are to be understood as the temperature settings for a given period of exposure to treatment. The critical temperature limits are set at a specific temperature or a temperature below that setting. Variance in the system (the regulations require that settings be set at a given value or below with no acceptance or allowance for temperatures above the specified setting) is controlled by monitoring the critical control limit. This implies that if a specific equipment has a variability of $\pm$ 0.1 degrees, the setting (critical control limit) required to achieve regulatory treatment for the sample combination "32^{o}F for ten days", will be 31.9^{o}F for ten days. The setting lower than 32^{o} F will allow for variability associated with specific equipment.

USDA maintains lists of approved vessels and approved cold treatment equipment that satisfies its requirements. These lists may be found in USDA (1998), updates to cold treatment recommendations can be obtained directly from USDA APHIS PPQ and are currently (2002) available directly from the USDA APHIS PPQ website ((http://www.aphis.usda.gov/oa/clementine/coldtxre.pdf). The USDA (1998) reference also includes descriptions of the requirements and instructions used to verify the accurate delivery and application of cold treatment.

Treatment schedules are designed to approximate a probit 9 (99.9968 percent) mortality and were based on a demonstrated large-scale confirmation tests that shows that the treatment kills about 100,000 insects with no survivors. The tests gives a statistical inference of a survivor rate of either 30 or 32 survivors or less with a binomial distribution with a mode of zero. This distribution does not allow for error in operational protocol which would have a mode greater than zero, nor does it consider that most treatments continue on ship even after the required number of days are completed, because the travel time from port to port is often greater than the required treatment time; this increases the mortality. (Liquido et al 1995; C.E. Miller pers. comm.).

Treatment schedules were based on probit 9 (99.9968 percent) mortality. This corresponds to a survival rate of 0.000032 (0.0032 percent). This distribution is consistent with the mortality patterns cited in the previous paragraph, and with characterization of insect treatment mortality in general (Robertson and Preisler, 1992).

The result from this step is that a proportion of at ca. 0.000032 larvae survive treatment. That value represents the proportion of survival for fruit flies. Supporting evidence for this value is provided by Back and Pemberton (1916a,b); Fares (1973); Flitters (1958); Hill et al. (1988); Mason and McBride (1934); Pettey and Griffiths (1931); Nel (1936). A year 2002 review of the evidence supporting cold treatments and the update of the cold treatment schedule is intended to provide additional safeguards on the existing cold treatment schedule (Gould *et al*., 2002) and to minimize variability about that value.

A bounded distribution (discrete minimums and maximums) was used to represent our assumptions of the variability about this component. Proposed changes to the treatment (increase treatment period by two days) are likely to increase mortality (bias the survivorship towards zero. We thus used a Pert distribution to simulate the survival of larvae with the following parameters: minimum of zero, most likely value of 0.000001 and maximum value of 0.00001.

Whereas there were suggestions (list of reviewers and comments presented at end of literature review section) that the variability in cold treatments may have been higher prior to implementation of increased vigilance measures by USDA and increased quality controls (i.e., during 2001 and before), we did not have sufficient evidence at the time of this writing to characterize this variability. We used the same parameters for the baseline and new program scenarios. We again note that recommendations in place after 2001 (e.g., Gould *et al*., 2002 and program workplan) are intended to reduce the variability in cold treatment and maximize efficacy. We also acknowledge that reviewers commented that observations during 2001 indicated less than probit-9 effectiveness associated with cold treatment. Table 1 (see end of table 1, columns titled "Ship surveillance 2001-Expected Value-Beta distribution parameters) was interpreted by a reviewer to confirm that with 2 "survivors" and a total of 210 larvae, the proportion that survived treatment was 9.52E-03 (Beta distribution parameters, including 95% lower confidence limit, lcl, and upper confidence limit, ucl, are reported in the table, as per suggestions by a reviewer). Clearly, this level of mortality is much higher than expected if we are to assume probit-9 effectiveness. We note however, that Baker et al. (1944) note that after cold exposure, some tephritid larvae (*Anastrepha ludens*) experience physiological dysfunction and do not lead to viable adults. This is also consistent with the research of Mason and McBride (1934). Still, Baker et al. note that a heartbeat continued to be measured for 47 days after treatment. This suggests that live larvae are not directly related to viable adults after exposure to cold treatment. Further evidence from 2001 (e.g., Administrative Record, pages 28-31) indicated that many of the larvae found were brownish in color or died hours after movement was observed. Healthy larvae are cream-colored but turn brownish when moribund or dead. It is clearly not practical to maintain rearing facilities at all inspection points and whereas this fact has led to a consistent policy of decision making based on live larvae found.

The actual effectiveness (e.g., probit-9 effectiveness) should be more appropriately linked to viability of resulting adults; such results are usually linked to experimentally derived laboratory observations, not to observations made at market places or ports of entry. Policies based on the finding of live larvae are based on the impossibility of locating laboratories or quarantine rearing facilities at all locations where interceptions may be made. These policies may be reviewed in the future if alternative, practical means of differentiating live larvae from moribund specimens become available.

Analysis completed in as part of USDA's review of risks associated with Spanish clementines includes USDA-ORACBA review of cold treatment data (2002; on the web at http://www.aphis.usda.gov/oa/clementine/coldtreatment7-5-02.pdf). This analysis confirms that previous treatment recommendations were not likely to provide mortality that is equivalent to probit-9. However, it also clearly notes that most treatments of more than 14 days in duration achieved mortality equivalent to, or greater than the benchmark probit-9. As per our parameters cited above, our analysis assumes that, especially given the more stringent controls and quality assurance associated with this pathway, cold treatment efficacy will be equivalent to probit-9, or better. Our analysis thus applies only to treatment schedules 14-days and above in duration or to other treatments that provide analogous levels of mortality (e.g., combinations of fumigation, cold treatment, and low prevalence-see USDA 1998 Treatment Manual).

Recent evidence cited during the public comment period and obtained as part of the review for this draft (e.g. De Lima et al. 2002) shows that large-scale tests in Australia confirm 100% mortality for treatments of greater than sixteen days duration. Whereas the entire range of temperature/duration schedules available in the United States was not tested by De Lima et al. 2002, they provide clear and conclusive evidence about the effectiveness of cold treatments for the treatments studied. Several other studies cited in the USDA-ORACBA review of cold treatment data (USDA 2002 Memo from M. Powell to D. Reeves; on the web at http://www.aphis.usda.gov/oa/clementine/coldtreatment7-5-02.pdf) confirm the effectiveness of cold treatments in terms of approximating the benchmark probit-9 level, or better.

Component 5 (C5). Arrives at suitable area (proportion of fruit discarded into suitable areas)

USDA (1993) has analyzed the portion of the United States at risk from *C. capitata* or the likelihood that a suitable host will be found in the southern portions of the continental United States. This incorporates both the likelihood that suitable hosts are in the area and the likelihood that an adult fly emerging from imported fruit will find the host material before dying.

Medfly is not likely to become established in an area where citrus does not grow. Areas that have winter temperature too cold for citrus are also too cold for the pest and citrus is generally the only good host available in subtropical or Mediterranean climates during the late winter or early spring (Miller 1992).

Fruit that arrives in the United States does not arrive at a single State. Rather, the fruit is distributed according to market demands through commercial distribution areas. The distribution channels and the fact that all fruit is destined for consumption reduce the number of fruit that end up in regions suitable to pests. U. S. demographics and distribution of markets are strong indicators of the ultimate destination of fruit. The distribution of U.S. population according to the 2000 U.S. Census is shown in figure 3 (http://www.census.gov/) and describes the likely patterns of fruit destined for human consumption. Fruit that enters is mostly directed away from suitable areas with a likely maximum 34 percent of imported fruit reaching states with citrus production. The US population varies between censuses and shows increasing trends towards higher densities in southern states. By 2025, the population that lives in the South and West (which includes states that have suitable conditions during most of the year; all areas of California, Arizona, New Mexico, Texas, Louisiana, Mississippi, Alabama, Georgia, Florida, and all US Islands and territories) may be 44% of the total US population (http://www.census.gov/). *We used 34 and 44% as minimum and maximum constant values, respectively.* Note that we do not imply here that the states considered have suitable conditions and susceptible hosts throughout the year; rather we used a conservative approach and focused on climatology. This was because of the uncertainty associated with the occurrence of susceptible hosts in several states (for example, New Mexico, Alabama, Mississippi; northern portions of California, Georgia, and Texas); we accounted for this uncertainty by assuming that all the southern states were climatologically suitable during all the year and that they had susceptible hosts.

The distribution of susceptible hosts does not cover an entire state for any given host or combination of hosts (http://www.usda.gov/nass/aggraphs/) (figure 4a-c); county level descriptions would be more appropriate. Such descriptions were not available at the time of this analysis.

We acknowledge that other methods have been used to assess suitable regions. Common alternatives involve predefined ecoregional divisions or U.S. "Plant Hardiness Zones" U.S. Department of Agriculture (e.g., USDA, 1990), our choice to combine both host suitability areas with population densities integrates the fact that markets are strong determinants of fruit distribution with climatic suitability. The areas (entire states) considered here are consistent with Smith (1993) but are also coarser in that areas smaller than one state are not distinguished. The impact of this difference is that our analysis will tend to overestimate areas suitable for Medfly establishment.

Comments received to earlier drafts of this document suggested that strains of Medfly in Spain might be considered cold tolerant and more likely to survive north of the citrus growing areas. This is certainly likely but is not supported by empirical evidence (no establishments of Medfly ever in areas of North America located north of citrus growing areas[citrus mentioned here as an indicator species, not to suggest that it is the only host]). Additionally, site visits could not confirm occurrence of Medfly in the interior regions in Spain, but rather gathered data that noted a distribution along the Mediterranean coasts. Indeed, the Medfly is reportedly common along the Mediterranean coast (Dominguez 1998). Dominguez (1998) stated that reports of Medfly from the interior (i.e., away from coastal areas) are largely due to the movement of produce from coastal areas. He noted that there are no damage reports from Castilla La Vieja and that the colder regions in the Central and inland portions of Spain are probably not suitable for the continuous presence of the Medfly. Finally, in the United States we have an indirect indicator of Medfly likelihood of establishment: *Anastrepha ludens*, the Mexican fruit fly or Mexfly. Comparison of *Anastrepha* to *Ceratitis* larvae at temperature slightly above freezing (1-3^{o} C) indicates that Medfly is more susceptible to cold temperatures than is the Mexfly (Baker et al., 1944). Nevertheless, despite the Mexfly's yearly occurrence in large numbers in Southern Texas in the past years, its similar dispersal capacity and the significant traffic of people and products from Texas to other states has not resulted in establishments of the Mexican fruit fly in states north of the citrus growing areas. We stress that we do not consider citrus alone as a likely host, simply an indicator species in the sense of Miller (1992). Additional empirical evidence is provided by the lack of Medfly outbreaks in non-citrus growing states. Additional empirical evidence is provided and by the fact that despite the occurrence of hosts and the importation of citrus fruit from Medfly infested areas, this pest has never established or had outbreaks recorded in Canada, where regulations against this pest are few.

Whereas the probability that a mated pair exists in a single container can be estimated, an entire container (some nine tons of produce) is very unlikely to be discarded and for its contents to be placed into conditions that might lead to a risky scenario (an infested fruit in contact with appropriate pupation site near a susceptible host, under proper environmental conditions). A reviewer to an earlier draft emphasized this fact and noted that what is important is the estimate of the actual produce that is likely to encounter a susceptible host. Research shows that in similar circumstances (shipments of fruit to market for consumption) the proportion of fruit that is not consumed and is discarded varies from 0.5% to 5%, with the latter value being a maximum estimate of fruit discarded after purchase (Wearing et al., 2001; Roberts et al. 1998). In this version and in response to the evidence, we have revised our estimate of the amount of fruit that actually represents a hazard (i.e., fruit that is not consumed and is discarded into a suitable environment).

We assumed that the maximum amount of fruit in a container to a given area that can pose risk is equivalent to 5% of the amount in given container, this is the maximum value cited in the evidence (Wearing et al., 2001; Roberts et al. 1998). We note that we further assumed that fruit that is discarded represents (in terms of Medfly infestations only) a population of fruit that is similar to that in the containers that originated them and that fruit from different containers were similar and likely to arrive at a suitable location.

Integrating the Components

The endpoint of the analysis provides information about the likelihood of introduction into suitable areas of the United States of fruit flies in containers of commercial clementines fruit from Spain. The assumption that a single container should be considered the main risk carrying unit has been advocated elsewhere (e.g., Landolt *et al*., 1984). We also assessed the likelihood that all containers in a year end up in suitable areas (a suitable area understood generally, as that where susceptible hosts occur) to provide a reference point. We consider that that latter scenario is based on conservative assumptions because fruit from containers would have to end up in close association with susceptible hosts and the susceptible host must be in the right phenological stage (e.g., have ripe fruit) and the conditions would have to be adequate. Relatively close association of discarded fruit with a susceptible host is necessary because fruit flies are reportedly poor fliers and natural spread is thus not expected to be great. Weems (1981) reports that spread occurs within one mile (1.6 Km) and Fletcher (1989) reports that spread may reach 12.5 miles (20 km). The dominant form of distribution of medfly has been human assisted movement (Smith, 1993).

The assumption that all containers are independent (and equally likely to lead to hazards and exposure) is conservative because many containers will be shipped to areas unsuitable for many reasons (no hosts, hosts with no fruit, temperatures that are too cold). Considering that the peak of exports occurs after the summer (even in Spain, medfly populations drop dramatically in the Fall and virtually disappear in Winter), many of the shipments are not likely to encounter ideal conditions or susceptible hosts. The assumption that a probability of fruit flies (a mating pair) associated with any of thousands of individual, independent containers is representative, is a conservative estimate. Additional future evidence may help remove this bias.

We combined the components (C2, C4) assuming that the attrition processes were independent, particularly the field treatments and the cold treatments. The number of live larvae (L) in containers over an entire year to suitable areas may be estimated by multiplication as follows:

$$L = C1 \cdot C2 \cdot C3 \cdot C4 \cdot C5 \qquad (1)$$

Where

C1 = Number of fruit shipped,
C2 = Fruit infested with larvae in the field,
C3 = Larvae per fruit, and
C4 = Cold treatment survival rate.
C5 = Suitable areas (discarded)

The probability of introduction is directly linked to the likelihood that a mated pair (one male and one female) will occur from a container of fruit. This probability has been studied by several researchers (Landolt *et al*. 1984; Baker et al, 1990; Mangan et al. 1997; Liquido et al. 1996; Liquido et al. 1997); key findings are applied here.

Vail et al (1993) has simplified the estimate to shown that the Probability, P of a mated pair in a shipment is defined as

$$P = [1-e^{-NR/2}]^2 . \qquad (2)$$

Where,

P is the probability of one or more mated pairs occurring in a given shipment
N is the number of fruit in a shipment (container),
R is the rate of infestation, and

e is the base of natural logarithms.

Note NR is simply the number of fruit infested with live larva in a shipment (container).

The infestation rate was estimated by dividing the fruit infested with live larva (NR) by the total number of fruit. Using our notation from above, R is defined as follows:

$$R = (C1 \cdot C2 \cdot C3 \cdot C4)/C1 = C2 \cdot C3 \cdot C4 \qquad (3)$$

Thus, we see the infestation rate is C2 (Fruit infested with larvae in the field), multiplied by C3, Larvae per fruit, multiplied by C4 (Cold treatment).

The probability of a mated pair in a single shipment (assumed to be destined to a suitable area) was estimated by the equation (2), with R from equation (3) above and using a value of N equal to the number of fruit in a container equal to C1(number of fruit). This is "result 1". We considered "result 1" as a key indicator and component of risk.

The probability of a mated pair in containers shipped to suitable areas over the entire year was estimated with equation (1) and, $1-(1-P)^S$ and we further assumed that 44% of containers (2820 per year) actually end up in suitable locations; additionally, five percent of fruit in those containers will be discarded ["result 2"]. We consider result 2 based on conservative assumptions as it is clear that if all shipments are independent and equally likely to encounter suitable conditions is a simplification. However, this simplification is needed due to our knowledge about the system and due to the necessary simplifications made by a model.

X. RESULTS

In order to assure replicability of our results and complete transparency of our calculations, the spreadsheet that details our estimations and presents all values explicitly is available from the technical contacts named at the end of this document and from the USDA APHIS website.

The input parameters and expected mean value of the components of the system are presented in Table 4a-c. The endpoints described above (results 1,2) are shown in table 4d.

Table 4d shows that the probability of a mated pair in a single shipment and under the mitigated (new program) scenario is lower than 0.000001 from "result 1". The probability that a mated pair arrives to a suitable location (in 2,820 containers shipped to suitable locations) is less than 0.0001 from "result 2". Previous versions of this analysis presented probabilities associated with containers shipped to all locations, independent of whether such locations had hosts or conditions suitable for Medfly. Those results have been removed because they are not a realistic representation of biology and because early reviewers found them confusing.

The variability associated with each component was explored using a Monte Carlo simulation procedure that combined all the possible values for all the components considered into an expression of overall probability and associated distribution of probable values. This combined probability then represents the overall pathway. In a typical Monte Carlo simulation, the endpoint value is calculated many times and is meant to produce a distribution of values, in addition to a single point estimate.

The risk analysis software package, @Risk™ for Excel was used to evaluate the effect of variability in the analysis of Medfly in fruit. Simulations for each component were run for 10,000 iterations. Input values for the calculations were drawn from the specified input distributions during each iteration (i.e., input values were drawn from values like the maximum and the minimums specified in table 4a and 4b), and the computer program randomly selected a value from each of the input distributions. After the specified number of iterations, the software generated a combined distribution, expressed in terms of the annual distribution of chances of the occurrence of mated pairs. Results of the simulation are summarized in Table 4d. The characterization of the variability is further discussed in Appendix 3.

The variability associated with the components did not change our assessment of the likelihood of entry of infested fruit from earlier risk assessments that proposed that cold treatment provided significant reductions in risks. That is, the level represented by the baseline values has been associated with an appropriate level of protection for two decades (that is, importation of Spanish clementines since early 1980s). However, we emphasize that this document does not assess what constitutes an appropriate level of protection; it merely points out that the results of the mitigation activities (Table 4d) further decrease the risks associated with the importation of clementines from Spain compared to the baseline.

The distributions chosen were Pert and Normal and were parameterized based on our knowledge of the maximum, minimum, most likely and other appropriate parameters that characterized the range of possible values. Our justification for using these distributions is that they capture the range and most expected values as indicated by our evidence (Vose 2000). Continuous distributions are appropriate approximations for discrete distributions only when very large numbers are involved (Steel and Torrie, 1980). Whereas we acknowledge that these distributions are not a 100% accurate reflection of underlying mechanisms, we are certain that we have captured the essential behavior of the system. Essential behavior in the sense that it is a portrayal of risks that is appropriate to support decision making because it captures the essential characteristics of the system, links scientific evidence to the different components of this system and expresses results using useful metrics.

The endpoint of our analysis showed that on average the probability that a pair of fruit flies occurs in shipments to the United States even if all containers shipped to suitable areas for an entire year are considered, is less than 0.0001. This document concluded that proposed new mitigation practices (notably, assuring low field populations of fruit flies) reduced overall risk compared to the current system of cold treatments alone.

Although this document addresses clementines specifically, the risk from other citrus from Spain may be comparable to that evaluated here because the pest complexes and risk mitigations practices are similar; however as a matter of policy, USDA will require new pest risk assessments for new commodity permit requests.

Shipments of clementines from Spain have been ongoing for some twenty years since the original decision was made to allow importations. That decision was based on the risk assessment methods in use at that time (that is, a decision sheet shown in appendix 4), which is equivalent to current procedures. In the intervening decades and using cold treatments alone, there have been no medfly outbreaks that can be attributed to Spanish clementines following commercial pathways. Further, twenty years of sampling these shipments at our ports of entry have provided us with data that suggests that overall this pathway is not consistent with high values of infestations or high likelihood of introduction of fruit flies. Our analysis confirms these empirical observations and notes that the addition of new phytosanitary measures (that is, limitations to the field densities of pests) will result in greater phytosanitary protection than the use of cold treatments alone.

During the development of this document, it was suggested by reviewers that our cold treatment assumptions [namely that probit-9 (or better) would be achieved by increased exposure and increased quality control] might be too narrow. Specifically, it was suggested that our analysis did not allow for failures in the system. We addressed potential failures in our system and failures in our assumptions in Table 5. The results in table five show that if we were to have a one order of magnitude drop in the cold treatment effectiveness (as characterized in table 4), we would observe the following: the probability of a mated pair in a container would be less than 0.001. The probability of mated pairs in multiple containers (to suitable areas only) would be ca. 0.2. The latter calculations assume that in addition to simultaneous, multiple failures, all containers encounter equally suitable environments such that emerging flies can find suitable hosts, suitable mates and mate.

Despite the examination of extremes above, the authors believe most uncertainty in this system (in the sense of Vose, 2000) is linked to variability, not to pure uncertainty; however, the effect of uncertainty was addressed by examination of hypothetical deviations of cold treatment effectiveness by up to one order of magnitude. Additional discussion of the treatment of variability and uncertainty is included in Appendix 3.

Chew (1996) noted that in some cases the use of probit-9 as indicative of quarantine security was not appropriate. More generally he state, "pre-set mortality rates also ignore several other factors: pre-shipment cultural practices, survival, and reproductive capacity of the organism; packaging and shipping conditions; seasonality of shipment; distribution of the commodity, etc." Especially in cases where "only sound fruit are shipped or if the fruit is a poor host" the probit-9 level of mortality may not be necessary. As Baker (1939) noted, "insofar as infestation can be detected, only sound fruit is admitted to shipment". Chew (1996) argues that the use of probit-9 by Baker in 1939 and the subsequent adoption of probit-9 as indicative of a pre-set level of security is confusing, because preset levels ignore the dynamic interplay between treatment mortality and other factors as identified above.

Our analysis acknowledges the value of cold treatment especially in the context of phytosanitary safeguards as represented in the workplan. It does not endorse fixed or pre-set levels of mortality as indicative of conditions that ensure that pests associated with a commodity do not pose risks. We agree with Chew (1996) that the level of mortality induced by a treatment and the overall probability of introduction question must be considered within the context of the overall pathway. We have attempted to do so in this document. Importantly, there is interplay between the two critical control points (low populations and proportion of survivors) as was noted by Chew (1996). The practical implication is that if future research can demonstrate improved effectiveness of quarantine treatments, the level of the initial infestation that will give analogous results to those studied here, may be higher. Simply put, if you kill a higher proportion with improved Treatment X, you can have a higher initial density compared to Treatment Y that kills a lower proportion of the exposed population. Clearly then, when using Treatment Y, the critical control limit (i.e., the threshold amount of allowable field densities) will be lower than when using Treatment X. This observation is an extension of Chew's remarks but it is an important reminder and consideration given the dynamics and evolutionary nature of phytosanitary treatments. As new research and methods development results are made available, the analysis presented here may be updated, as appropriate.

XI. LIKELIHOOD OF INTRODUCTION

The analysis in this document has emphasized the calculation of infested fruit, Medflies in fruit, and probability of a mated pair; however, once a mated pair arrives it must still overcome physical and biological hurdles before it becomes established and spreads.

There are additional mitigations which provide safeguards but which were not evaluated quantitatively but are described qualitatively. A key component of such additional safeguards is the rejection of infested lots found by port inspections. Port inspections, especially for citrus are being revised and updated at the time of this analysis. The intent of the revision is to increase the safeguarding value of port inspections. Whereas we do not present quantitative values for this component at this time, we state qualitatively that port inspections are an additional mitigation measure of significant value.

As noted, the endpoint of this assessment is not probability of introduction but probability of a mated pair arriving in containers shipped to the United States. Fruit flies must survive refrigerated storage, emerge from fruit onto suitable soil to pupate; escape predation, emerge from pupation, find a sexually mature mate, mate, find suitable environment, find a host, find fruit that is sufficiently mature, oviposit viable eggs, avoid death by desiccation/heat/cold, and others (Light and Jang, 1995; Bateman, 1972).

Another element that makes this estimate conservative is that we used demographic projections 25 years into the future as the basis for our calculations. The expectation in 25 years is that there will be larger markets (by virtue of population migration and other demographic dynamics) in areas where suitable conditions and hosts occur ("the south"). This implies that present day risks are lower.

The above factors were not evaluated quantitatively but are significant in terms of understanding why this commercial, monitored, treated pathway of fruit for consumption and distribution through US market channels is consistent with low likelihood of introduction of Mediterranean fruit flies. Finally, the timing of imports from Spain is biologically significant. Imports occur towards the beginning of the cooler months of Fall and Winter (Shipments from Spain normally occur in September-December), a time when the potential for suitable conditions in most of the United States is decreased due to lower temperatures and decreased presence of fruit hosts (especially after November). The result of the lack of information regarding the value of the mitigations above is that this assessment is not unbiased, it is conservative. Additional future information will allow us to remove some of the bias in the present analysis; however, it is clear that considering the above our results provide strong evidence that the likelihood of introduction of Mediterranean fruit flies along the commercial, mitigated, monitored pathway is very unlikely and insignificant in terms of traditional statistical measures that suggest that probabilities lower than 0.0001 in biology usually translate to "very rare events," especially when compared to known baseline hazard exposures (e.g., hundreds of fruit flies arrive each year in passenger baggage). It is in that sense that the term "insignificant" is applied.

We note that although we did not evaluate the likelihood of introduction quantitatively, we do know the value of its upper bound. The maximum value of likelihood of introduction is the probability that a mated pair occurs in shipments of clementines (the values estimated in this article and summarized in table 4d). The reason that this is a maximum value is because other processes to be considered in establishing likelihood of introduction (and assuming that the United States continues to be an area free of Medflies) will only result in attrition or death processes. That is, after a potential fruit fly arrives in a container of fruit, the additional hurdles it must overcome will result in reductions in its likelihood to become successfully introduced.

XII. CONCLUSIONS

Our analysis shows that the combination of population control in production fields combined with effective application of cold treatments (as updated during 2002), results in reduced risks compared to the use of quarantine cold treatments alone. Previously other citrus fruit from Spain has been allowed entry into the United States with cold treatment for Medfly. These include sweet oranges (*Citrus sinensis)*, other varieties of *Citrus reticulata,* ortaniques *(Citrus sinensis x Citrus reticulata),* and ethrogs (*Citrus medica*). Lemon, sour limes and under certain conditions, ethrogs, are allowed entry without treatment because of non-host status. Although this document addresses clementines specifically, the risk from other Medfly host citrus from Spain may be comparable. The other fruit are similar or larger thus, less fruit would be in the shipment and the number of pests per shipment may be similar.

The critical control points -cold treatment and field population control- are being addressed by both existing (e.g., USDA, 1998 Treatment Manual) and new procedures (workplan; Gould *et al*., 2002). These control procedures will assure that the risk mitigations will be maintained as evaluated in this document.

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Review and comments received through June 2002 (a full listing of those that provided input through the end of the comment period on September 9, 2002 can be found at www.aphis.usda.gov)
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The above list identifies early input or comments received before the completion of the final version of this document. Additional comments were addressed and evaluations updated in this version (September 2002). A full listing of public comments and stakeholders is part of the public record.

We made changes to this document in response to comments received on earlier drafts as follows:

1. We corrected inconsistencies between text and tables.
2. We added new evidence to support values used, corrected distributions. Specifically, added data on direct observations from ships as fruit was unloaded at ports of entry and from distribution outlets. This direct sampling data, especially given the large sample sizes are the best indicator of presence of infestations in the fruit. Three new detailed tables summarize this evidence.

3. We included corrections to account for reduced reliability of sampling; specifically added evidence about the proportion of infested fruit that are likely to be detected and then corrected our observations to consider the partial efficiency of inspectors.

4. In order to further investigate uncertainty, we decreased the efficiency of the cold treatment 10-fold and presented further discussions on uncertainty and variability.

5. We added new evidence that the range of the fly has not extended into colder areas of Europe and evidence that more cold tolerant fruit flies present in the South of Texas have not migrated or established in northern US.

6. Public comments about the mathematical treatment of the data, the science behind the assessment, the validity, of parameters, the validity of assumptions, the validity of the model, and the validity of the data were addressed by adding more data and direct observation evidence to the analysis, by modifying the model to ensure that model components were independent, by demonstrating low incidence of pests in fruit with observations directly from field sampling, and by adding new evidence on the survival from cold treatment that suggests that live larvae found after treatment are often not viable and evidence about cold treatment effectiveness as per assumptions in the analysis.

7. Transparency and complexity of the model used were increased by adding more discussion and examples and by providing the actual spreadsheets and all calculations used to all requestors. These documents were also provided on the USDA APHIS PPQ Internet website.

An early version of this draft was distributed via the USDA APHIS PPQ website. This version represents our final draft, September 2002 and includes comments and refinements in response to all comments and evidence received. Additional response to comments was produced in separate documents published as part of the proposed rule. The "response to comments" documents released as part of the rule making process are included here by reference.

XIV. TEAM

The following PPQ groups participated in the development or provided input for this document: CPHST, PPD, IS, PIM, ARS, Regions and ports (especially Port of Philadelphia), SITC. Additional information was received from IPPC and the Spanish Ministry of Agriculture, Fisheries, and Nutrition.

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TABLE 1. FRUIT SAMPLED FOR FRUIT FLY; SAMPLES FROM VESSELS IN 2001
Source: USDA-APHIS-PPQ Sampling.

Vessel Name	No. Fruit cut	No. Dead Larvae	No. Live Larvae	Total Larvae	Brand Name
Greenwich Maersk	50	0	0	0	Tina
S/L Performance	50	0	0	0	Bombi
S/L Performance	50	0	0	0	AMC
Greenwich Maersk	50	0	0	0	Tina
S/L Performance	50	0	0	0	Dia Sol
S/L Performance	50	0	0	0	Dia Sol
Greenwich Maersk	50	0	0	0	Bombi
Margrethe Maersk	50	0	0	0	Falcon
S/L Performance	50	0	0	0	Bombi
Crown Topaz	180	0	0	0	Blink
Crown Topaz	60	0	0	0	Deica
M/V Interray	180	0	0	0	Flamenco
Crown Topaz	60	0	0	0	Blink
Polar Argentina	120	0	0	0	Blink
S/L Commitment	60	0	0	0	Gourmano
Polar Argentina	60	0	0	0	Peica
S/L Quanlity	150	0	0	0	SIPS
S/L Quanlity	150	0	0	0	Superior
S/L Quanlity	150	0	0	0	Sweetie
S/L Quanlity	150	0	0	0	SIPS
S/L Quanlity	150	0	0	0	Superior
S/L Quanlity	150	0	0	0	Dia Sol
S/L Quanlity	150	0	0	0	Superior
S/L Quanlity	150	0	0	0	Sweetie
S/L Quanlity	150	0	0	0	Superior
S/L Quality	150	0	0	0	SIPS
M/V Marstal Maersk	150	0	0	0	Gourmand
M/V Marstal Maersk	150	0	0	0	Tono
S/L Quality	150	0	0	0	SIPS
S/L Integrity	150	0	0	0	Camalu
S/L Quality	150	0	0	0	Bombi
S/L Quality	150	0	0	0	Spanish
M/V Marstal Maersk	150	0	0	0	AMC
S/L Quality	150	0	0	0	AMC
S/L Quality	150	0	0	0	Dia Sol
S/L Performance	150	0	0	0	AMC
M/V Marstal Maersk	150	0	0	0	AMC
S/L Quality	150	0	0	0	AMC
S/L Performance	150	0	0	0	AMC
S/L Quality	150	0	0	0	AMC
S/L Quality	150	0	0	0	Dia Sol
S/L Quality	150	0	0	0	Sweetie
S/L Quality	150	0	0	0	Sweetie
S/L Quality	150	0	0	0	Dia Sol
Italian Reefer	1490	0	0	0	unknown
Green Malloy	1320	0	0	0	Bicoca
Green Malloy	20	0	0	0	Sabrosa

Table 1 (continued).

Vessel Name	No. Fruit cut	No. Dead Larvae	No. Live Larvae	Total Larvae	Brand Name
M/V Interray	240	1	0	1	Camalu
S/L Commitment	60	1	0	1	Sweetie
S/L Commitment	60	1	0	1	Sweetie
Margrethe Maersk	150	1	0	1	Falcoln
S/L Commitment	60	2	0	2	Sweetie
M/V Performance	50	2	0	2	Bombi
American Eurost	1490	2	0	2	Desin Nature
S/L Commitment	60	3	0	3	Sweetie
S/L Performance	120	3	0	3	Sweetie
S/L Quanlity	150	3	0	3	Green Time
Polar Chile	120	3	0	3	La Rica
M/V Performance	50	3	0	3	Bombi
M/V Hamburg	1490	3	0	3	Darling
M/V Performance	50	4	0	4	Bombi
M/V Marstal Maersk	150	2	2	4	Tina
Baltic Snow	1490	4	0	4	Badilis
Baltic Snow	1490	4	0	4	Ocean Spray
Polar Argentina	60	5	0	5	Diana
M/V Performance	50	6	0	6	Fruitisol
S/L Quality	150	6	0	6	AMC
M/V Interray	120	7	0	7	Diana
S/L Quality	150	7	0	7	AMC
S/L Performance	240	8	0	8	Sweetie
M/V Marstal Maersk	150	8	0	8	Tono
Baltic Snow	1490	8	0	8	Bagus
Crown Topaz	120	10	0	10	Tienta
Amer Everest	240	10	0	10	Clementines
Crown Topaz	120	12	0	12	Tienta
S/L Quality	150	12	0	12	Bombi
S/L Commitment	120	15	0	15	Gourmano
Baltic Snow	1490	16	0	16	Nadal
M/V Marstal Maersk	150	18	0	18	AMC
Asian Reefer	180	20	0	20	NX
Totals	20460	210	2	212	

Table 1 (continued).

Ship Surveillance 2001		**Expected Value**	**Beta distribution parameters***				
Survivors**	**total**	**Prop. survive**	**s^2**	**alpha**	**beta**	**lcl**	**ucl**
2	210	9.52E-03	4.51E-05	1.98	206.02	0.001	0.026

Infested Fruit	**Total fruit**	**Infested fruit/total**	**s^2**	**alpha**	**beta**	**lcl**	**ucl**
33	20460	0.00161	7.87E-08	33	20425	0.001	0.002

*Beta distribution parameters obtained by the method of matching moments (Evans *et a*l., 1993).
** Survivors refer to live larvae, it is however not known whether these live larvae were viable. Evidence presented in the text suggests that after cold treatment, many larvae survive (some for weeks) but are unable to produce viable adults.

average larvae/ infested fruit	**std**	**95% confidence**
6.4	5.16	1.76

TABLE 2. FRUIT SAMPLED FOR FRUIT FLY FROM OUTLETS (MARKETS), SEVERAL LOCATIONS, 2001
Source: USDA-APHIS-PPQ Sampling

Date	City/State	Fruit Cut	Dead Larvae	Live Larvae	Total Larvae	Brand Name
30-Nov-01	Baton Rouge, LA	3	0	0	0	Darling
30-Nov-01	Baton Rouge, LA	4	0	0	0	Maxim
1-Dec-01	Chula Vista, CA	15	0	0	0	Filosofo,Elite
1-Dec-01	Coronado, CA	15	0	0	0	Filosofo, Elite
1-Dec-01	La Jolla, CA	20	0	0	0	Llusar
1-Dec-01	La Jolla, CA	15	0	0	0	Evyan, Filosofo
1-Dec-01	San Diego, CA	10	0	0	0	Elite
1-Dec-01	San Diego, CA	15	0	0	0	Filosofo
1-Dec-01	San Diego, CA	10	0	0	0	Elite
1-Dec-01	San Diego, CA	10	0	0	0	Elite
1-Dec-01	San Diego, CA	10	0	0	0	Elite
1-Dec-01	San Diego, CA	15	0	0	0	Filosofo
1-Dec-01	San Diego, CA	10	0	0	0	Filosofo
1-Dec-01	San Diego, CA	15	0	0	0	Filosofo
1-Dec-01	San Diego, CA	15	0	0	0	Llusar, Elite
1-Dec-01	San Diego, CA	25	0	0	0	Filosofo
1-Dec-01	San Diego, CA	25	0	0	0	Evyan
1-Dec-01	San Diego, CA	10	0	0	0	Filosofo
1-Dec-01	San Diego, CA	10	0	0	0	Filosofo
3-Dec-01	Sapalpa, OK	3	8	0	8	Garcia, Ballaster
4-Dec-01	Webster Grove, MO	10	11	0	11	Del Monte
4-Dec-01	Oklahoma City, OK	7	13	0	13	Tropicana
4-Dec-01	Oklahoma City, OK	2	0	0	0	Tropicana
4-Dec-01	Lakeside, CA	25	0	0	0	Elite
4-Dec-01	Lakeside, CA	35	0	0	0	Llusar
4-Dec-01	San Diego, CA	40	0	0	0	Filosofo
4-Dec-01	Belle Chasse, LA	72	0	0	0	Del Monte
4-Dec-01	Belle Chasse, LA	144	9	0	9	Maxim
4-Dec-01	Houma, LA	20	0	0	0	Garcia, Ballester
4-Dec-01	Houma, LA	15	0	0	0	Llusas
4-Dec-01	Shreveport, LA	22	0	8	8	Evyan
4-Dec-01	Morgan City, LA	48	0	0	0	Darling, NX
4-Dec-01	New Iberia, LA	24	0	0	0	Darling, NX
4-Dec-01	New Iberia, LA	24	0	0	0	Del Monte
4-Dec-01	New Iberia, LA	12	0	0	0	Bagu
4-Dec-01	Abbeville, LA	24	0	0	0	Darling, NX

Table 2. (cont…)

Date	City/State	Fruit cut	Dead Larvae	Live Larvae	Total Larvae	Brand Name
4-Dec-01	Rayne, LA	4	0	0	0	Del Monte
4-Dec-01	Lafayette, LA	4	0	0	0	Maxim
4-Dec-01	Lake Charles, LA	6	0	0	0	Maxim
4-Dec-01	Lake Charles	6	0	0	0	NX
5-Dec-01	Baton Rouge, LA	5	0	0	0	Superior
5-Dec-01	Baton Rouge, LA	3	0	0	0	Tina
5-Dec-01	Baton Rouge, LA	5	0	0	0	NX
5-Dec-01	Gonzales, LA	5	0	0	0	Maxim
5-Dec-01	Gonzales, LA	6	0	0	0	Sealed Sweet
5-Dec-01	Galleano, LA	15	0	0	0	NX
5-Dec-01	Galleano, LA	15	0	0	0	Garcia, Ballester
5-Dec-01	Galleano, LA	15	0	0	0	Darling
5-Dec-01	Thibidaux, LA	30	0	0	0	NX
5-Dec-01	Lake Charles, LA	10	0	0	0	Soald Sweet
5-Dec-01	Lake Charles, LA	6	0	0	0	Blue Planet
5-Dec-01	Lake Charles, LA	4	0	0	0	NX
5-Dec-01	Alexandria, LA	12	0	0	0	Maxim
5-Dec-01	Natchitoches, LA	10	0	0	0	Evyan
5-Dec-01	Natchitoches, LA	6	0	0	0	Evyan
5-Dec-01	Natchitoches, LA	4	0	0	0	Llusas
5-Dec-01	Alexandria, LA	10	0	0	0	NX
5-Dec-01	Hartford, Ct	4	0	0	0	Sabrosa Golden Garden
5-Dec-01	New Haven, CT	9	0	0	0	Falcoln
5-Dec-01	East Haven, CT	11	0	0	0	NX
5-Dec-01	New Haven, CT	7	0	0	0	SIPS
5-Dec-01	New Haven, CT	4	0	0	0	ASI
5-Dec-01	Summit Point, WV	50	0	0	0	Peica
5-Dec-01	Summit Point, WV	55	0	0	0	Happy Tree
5-Dec-01	Waterbury, CT	12	6	0	6	Roxy
5-Dec-01	Waterbury, CT	10	0	0	0	Tina
5-Dec-01	Waterbury, CT	10	0	0	0	Del Monte
5-Dec-01	Waterbury, CT	20	10	0	10	Dolcita
5-Dec-01	Waterbury, CT	4	0	0	0	Roxy
5-Dec-01	Waterbury, CT	5	5	0	5	Flamerco
5-Dec-01	Waterbury, CT	6	0	0	0	AMC
5-Dec-01	Waterbury, CT	10	0	0	0	Three Sisters
5-Dec-01	Waterbury, CT	12	0	0	0	Del Monte
5-Dec-01	Waterbury, CT	8	0	0	0	Bagus

Table 2. (cont…)

Date	**City/State**	**Fruit cut**	**Dead Larvae**	**Live Larvae**	**Total Larvae**	**Brand Name**
5-Dec-01	Waterbury, CT	1	0	0	0	Sabrosa
5-Dec-01	Waterbury, CT	2	0	0	0	Roxy
5-Dec-01	Oklahoma City, OK	23	13	0	13	Superior
5-Dec-01	St. Louis, MO	6	0	0	0	Nimbus
5-Dec-01	St. Louis, MO	8	0	0	0	Darling
5-Dec-01	St. Louis, MO	6	0	0	0	Tienta
5-Dec-01	St. Louis, MO	8	0	0	0	Nimbus
5-Dec-01	St. Louis, MO	11	0	0	0	Bru Bru
5-Dec-01	St. Louis, MO	8	0	0	0	Falcon
6-Dec-01	Florissant, MO	8	0	0	0	Del Monte
6-Dec-01	Florissant, MO	8	0	0	0	Bru Bru
6-Dec-01	Florissant, MO	8	0	0	0	Darling
6-Dec-01	Florissant, MO	8	0	0	0	Falcon
7-Dec-01		12	0	0	0	
7-Dec-01		100	0	0	0	
7-Dec-01		60	0	0	0	
7-Dec-01	Herndon, VA	8	0	0	0	AMC
11-Dec-01	Madison, WI	24	0	0	0	Blue Planet Darling
11-Dec-01	Frankfort, IL	15	0	0	0	
12-Dec-01	Atlanta, GA	10	0	0	0	Del Monte
12-Dec-01	Atlanta, GA	10	0	0	0	Superior
12-Dec-01	Atlanta, GA	10	0	0	0	Fontcoop
12-Dec-01	Atlanta, GA	10	0	0	0	Tropicana Premium
12-Dec-01	Atlanta, GA	10	0	0	0	Golden Garden
1-Dec-01	San Diego, CA	10	0	0	0	Elite
1-Dec-01	San Diego, CA	5	0	0	0	Filosofo
1-Dec-01	San Diego, CA	20	0	0	0	Llusar
1-Dec-01	San Diego, CA	12	0	0	0	Bagu
1-Dec-01	Imperial Beach, CA	25	0	0	0	Elite
1-Dec-01	San Diego, CA	10	0	0	0	Bagu
3-Dec-01	San Diego, CA	12,966	0	0	0	
3-Dec-01	San Diego, CA	1,170	0	0	0	
3-Dec-01	San Diego, CA	660	0	0	0	
3-Dec-01	San Diego, CA	2,478	0	0	0	

Table 2. (cont…)

inf. Fruit	total fruit sampled	Proportion Infested (infested fruit/total fruit sampled)	s^2	Alpha	Beta	Lcl	Ucl
9	18927	0.00048	2.51E-08	9.0	18916	0.0002	0.0008
		Average larvae/ infested fruit	**std**	**95% confidence**			
		9.2	2.82	1.84			

Table 3a. Fruit sampled from distribution outlets, California 12/01/01 to 12/11/01
Source: Joint Federal and State response team, reported by J. Goode, USDA-APHIS-PPQ

Number Fruit		Live larvae-single fruit	Dead larvae-single fruit	
2323		0	0	
180		0	0	
20		0	0	
1180		0	1	
100		0	1	
780		0	2	
150		0	2	
144		0	2	
405		0	3	
220		0	3	
1600		0	7	
1040		0	9	
860		0	13	
960		0	17	
1240		0	25	
	sum			**sum**
	11202			85

average larvae per fruit	average fruit infested	average larvae per infested fruit	95%Confidence
0.0076	0.00107	7.08	4.3

Table 3b. Fruit Fly Samples from distribution outlets (markets), California, December 2001
Source: J. Hillard (California Dept. of Food and Agriculture), only positive samples shown, total fruit not recorded.

Live larvae (single fruit)	**Dead larvae- single fruit***	**Total Larvae per fruit**
4	NR	4
1	NR	1
1	NR	1
1	NR	1
1	NR	1
3	NR	3
0	4	4
0	25	25
0	9	9
0	13	13
0	2	2
0	22	22
0	8	8
6	15	21
0	10	10
0	1	1
0	44	44
0	7	7
0	4	4
0	9	9
0	3	3
0	8	8
0	3	3
0	10	10
0	4	4

average larvae/ infested fruit	**95%Confidence**
8.72	3.914234151

*NR is not recorded.

Table 4a. Values used in the Estimation of *Mitigated* Risk*

C1 **Number of Fruit per container**	C2 **Fruit Infested**	C3 **Larvae per fruit**	C4 **Cold treatment Survivors**	C5 **Reaches Suitable area * Discarded Proportion**
Mean 166,050; (6408 containers per year)	Maximum 0.015	Maximum 8	Maximum 0.00001	Maximum 0.44 * 0.05
	Minimum 0	Minimum 1	Minimum 0	
Std. Dev. 15,375	Most Likely 0.001	Most Likely 3	Most Likely 0.000001	Minimum 0.34 * 0.05
Distribution: Normal	Distribution: Pert	Distribution: Pert	Distribution: Pert	Constant

Table 4b. Values used in the Estimation of Baseline Risk

C1 **Number of Fruit per container**	C2 **Fruit Infested**	C3 **Larvae per fruit**	C4 **Cold treatment Survivors**	C5 **Reaches Suitable Area * Discarded Proportion**
Mean 166,050; (6408 containers per year)	Maximum 0.15	Maximum 8	Maximum 0.00001	Maximum 0.44 * 0.05
	Minimum 0	Minimum 1	Minimum 0	
Std. Dev. 15,375	Most Likely 0.001	Most Likely 3	Most Likely 0.000001	Minimum 0.34 * 0.05
Distribution: Normal	Distribution: Pert	Distribution: Pert	Distribution: Pert	Constants

Table 4c. Mean (and 95th percentile) values of components evaluated for Spanish clementines imports

Parameter/ **Scenario**	**C1** **Number of Fruit Shipped**	C2 **Fruit Infested**	**C3** **Larvae per fruit**	**C4** **Cold treatment**	**C5*** **Reaches Suitable Area * Discarded Proportion**
Fruit Flies Mitigated	166,049 (191,325)	0.003 (0.008)	4 (6)	2.3E-06 (5.4E-06)	0.02
Fruit flies Baseline	166,051 (191338)	0.02 (0.07)	4 (6)	2.3E-06 (5.4E-06)	0.02

C5 is used to determine the proportion of all containers that are exposed (by virtue of not being consumed and being discarded) such that they constitute a hazard proper (most fruit are consumed and do not constitute a hazard).

Table 4d. Evaluation of components for Spanish clementines imports, mean (and 95th percentile)*

Scenario	**P [Mated Pair] per container[/1] (result 1)**	**$P_{multiple}$[Mated Pair] suitable locations (result 2)**
Mitigated (field controls plus cold)	Less than 1E-06 (1.4E-07)	Less than 1E-04 (0.0004)
Baseline (cold only)	Less than 0.0001 (9.3E-06)	Less than 0.001 (0.03)

/1 N.B. (these table footnotes are summarized from the text):

P[Mated Pair]per container = $[1-e^{-NR/2}]^2$.
where, P= probability of a Mated Pair per container, N = number of fruit, NR is the number of fruit infested with live larva in a container.

The infestation rate was estimated by dividing the fruit infested with live larva (NR) by the total number of fruit. Using our notation from above, R is defined as follows: $R = (C1 \cdot C2 \cdot C3 \cdot C4)/C1 = C2 \cdot C3 \cdot C4$

The estimation of the probability of at least one mated pair in multiple containers ($P_{multiple}$) was estimated as $P_{multiple} = 1-(1-P)^S$, where P is the probability of a mated pair in one container and S is the number of containers shipped to suitable locations (S = 6408 multiplied by C5 equals 141); all 95th percentile values shown were obtained from simulation.

The total containers (6408) were estimated from maximum estimates (106,406 metric tons per year) divided by the number of fruit per container.

*Additional details and uncertainty analysis in Appendix 3 and electronic spreadsheet (spreadsheet available from technical contact and from USDA APHIS PPQ website). Note that table 4a and 4b differ only in the value of the maximum for C2. Note too, that a "less than" format is used to refer to the mean, actual values shown in Appendix 3.

Table 5a. Values used in the estimation of extreme ("failures") values likelihood of mated pairs*

C1	C2	C3	C4	C5
Number of Fruit Shipped	**Fruit Infested**	**Larvae per fruit**	**Cold treatment Survivors**	**Reaches Suitable area**
Mean 166,050; (6408 shipments per year)	Maximum 0.15	Maximum 30	Maximum 0.0001	Maximum 0.7
	Minimum 0	Minimum 1	Minimum 0	Minimum 0.34
Std. Dev. 15,375	Most Likely 0.001	Most Likely 10	Most Likely 0.00001	Most Likely 0.4
Distribution: Normal	Distribution Pert	Distribution Pert	Distribution Pert	Distribution Pert

Table 5b. Expected values of components for Spanish clementines imports given assumptions with extreme values, mean (and 95th percentiles)

Parameter/ Scenario	**C1** **Number of Fruit**	C2 **Fruit Infested**	**C3** **Larvae per fruit**	**C4** **Cold treatment**	**C5** **Reaches suitable area**
Fruit Flies Extreme values, "failures"	166,050 fruit (191,336)	0.03 (0.07)	4 (6)	2.3E-05 (5.4E-05)	0.02

Table 5c. Evaluation of components for Spanish clementines imports assuming extreme values "Failures", values are mean (and 95th percentiles)

Scenario	**P [Mated Pair] Single container**	**P [Mated Pair], multiple containers**
Failures, Extreme Values	Less than 0.001 (0.001)	0.2 ~1

**These tables (5a-c) represent hypothetical failures in the system and extreme values chosen to investigate the behaviour of the system beyond the evidence presented.

Figure 1. Commercial citrus production areas in Spain (yellow), numbers indicate approximate total area for a region.

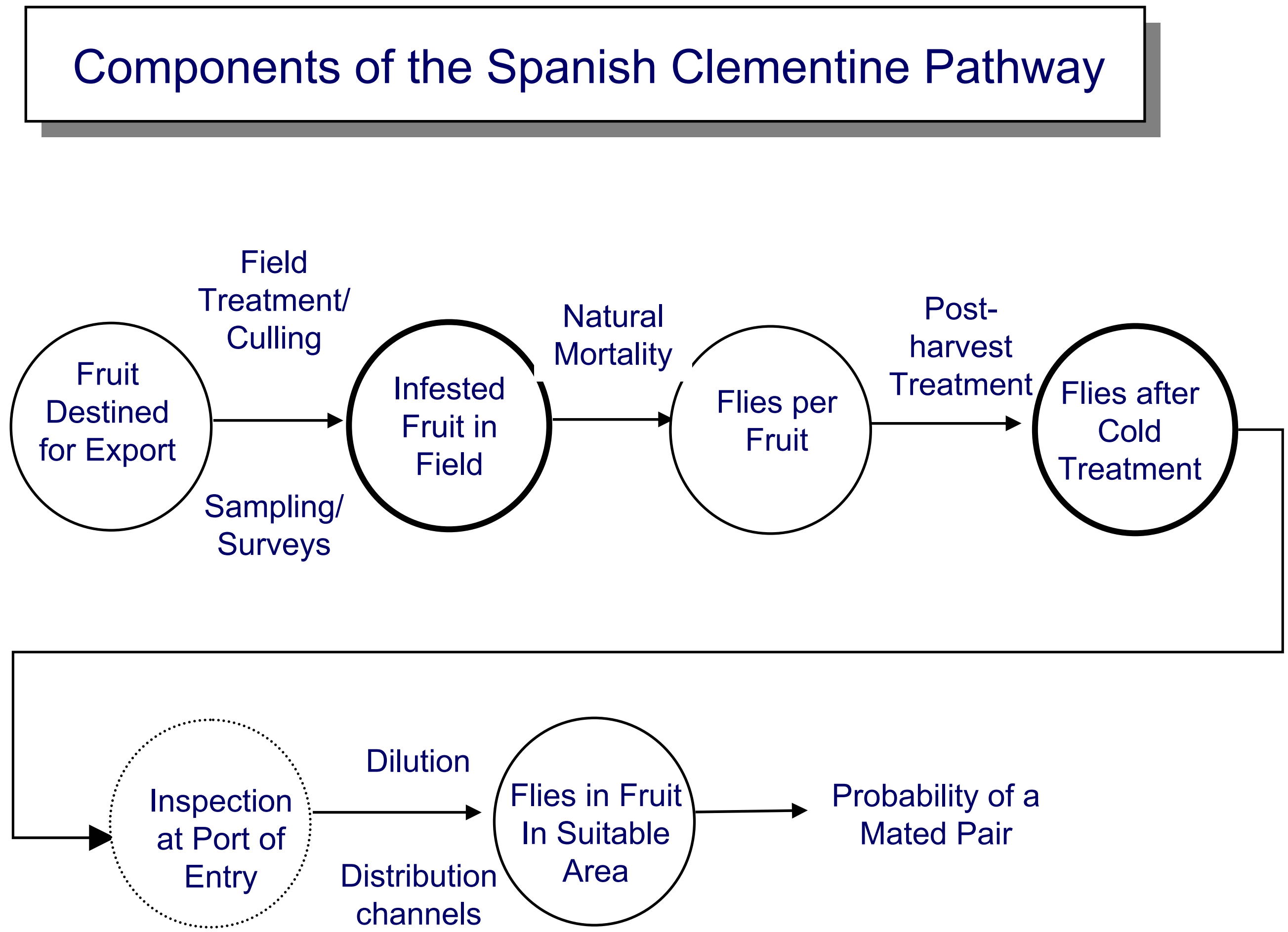


Figure 2. Components of the citrus pathway, bolded circles indicate critical control points, dashed circles indicate a mitigation component that was not evaluated due to incomplete data.

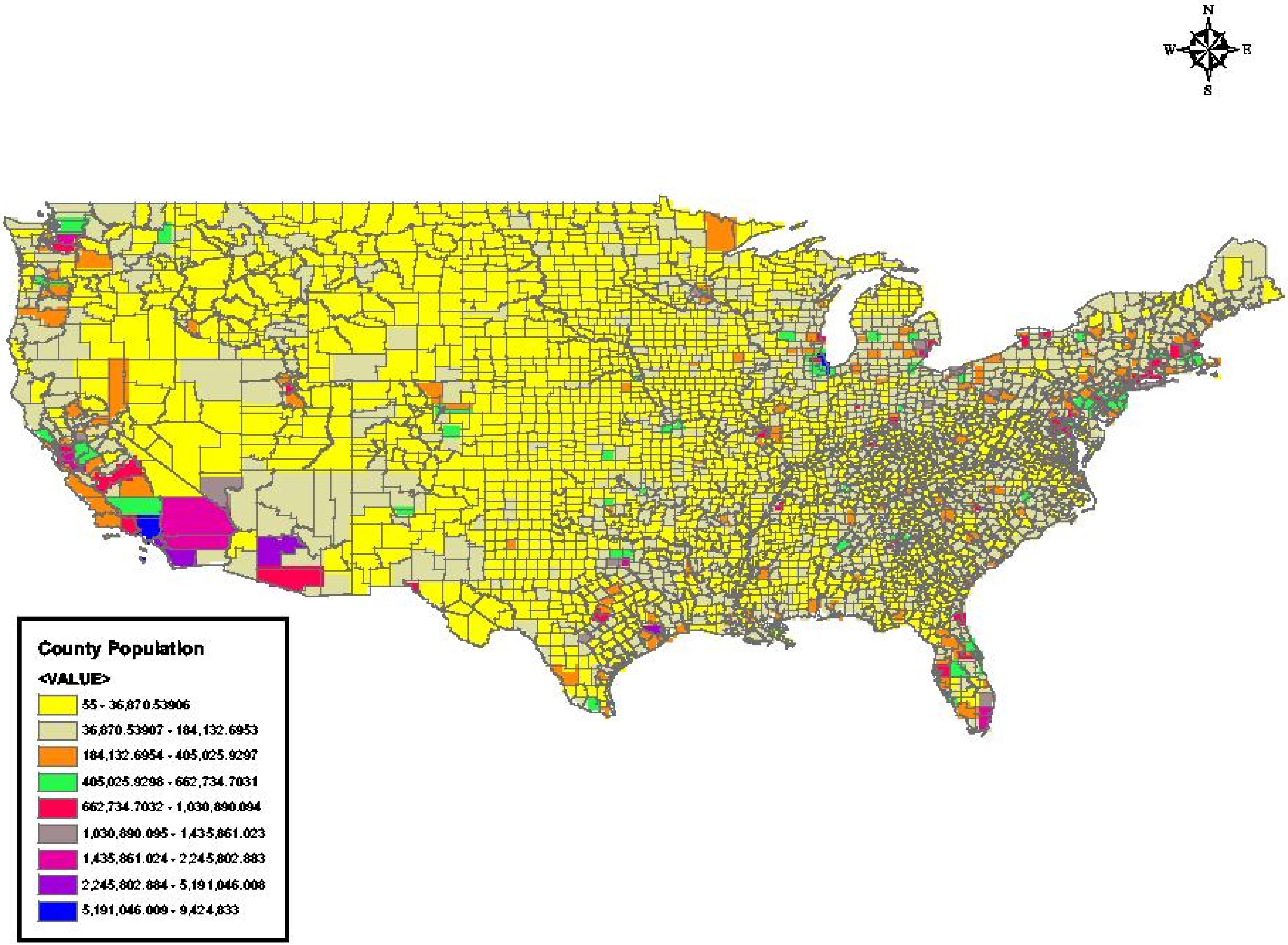
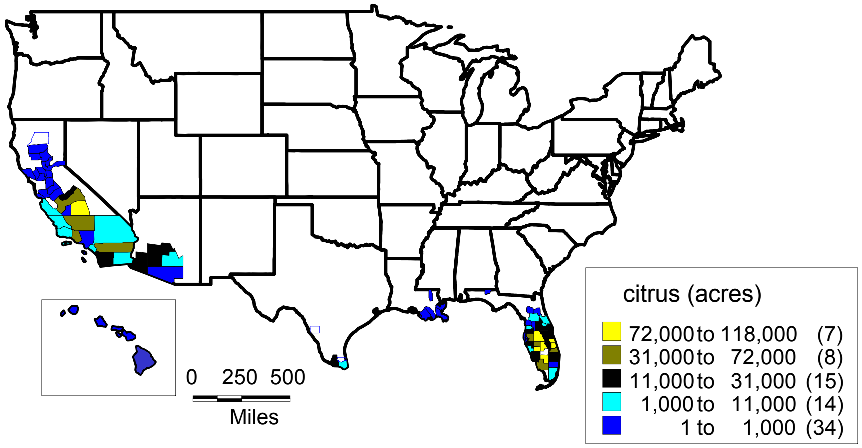


Figure 3. US population density, by county (US Census 2000)

Citrus



Mango

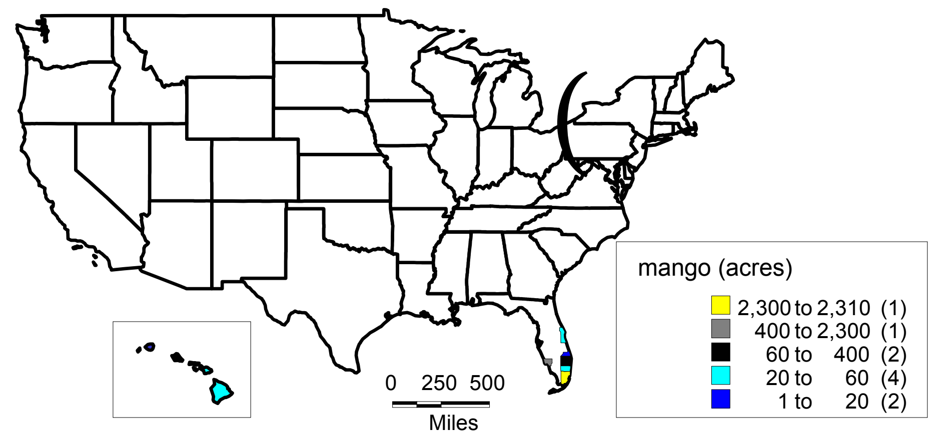


Figure 4b. Distribution of mango grove acreage in the United States (all commercial cultivars).

Figure 4a. Distribution of Citrus grove acreage in the United States (all commercial species and cultivars).

Figs

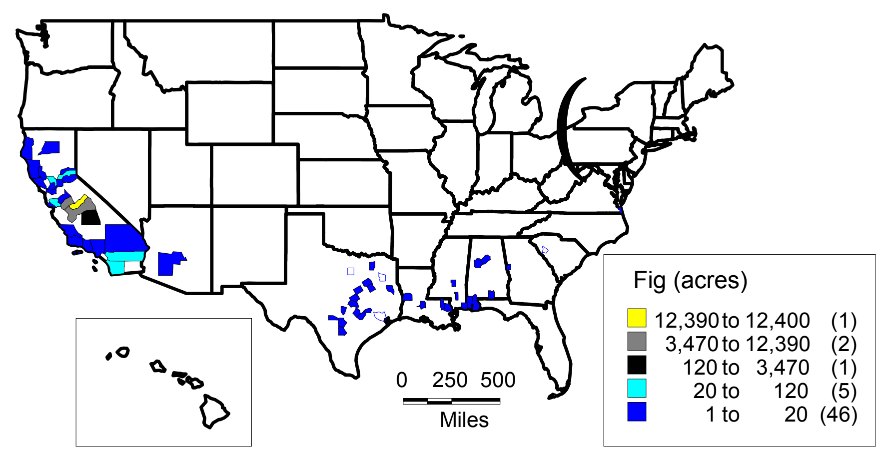


Figure 4d. Distribution of fig orchard acreage in the United States (all commercial cultivars).

Avocado

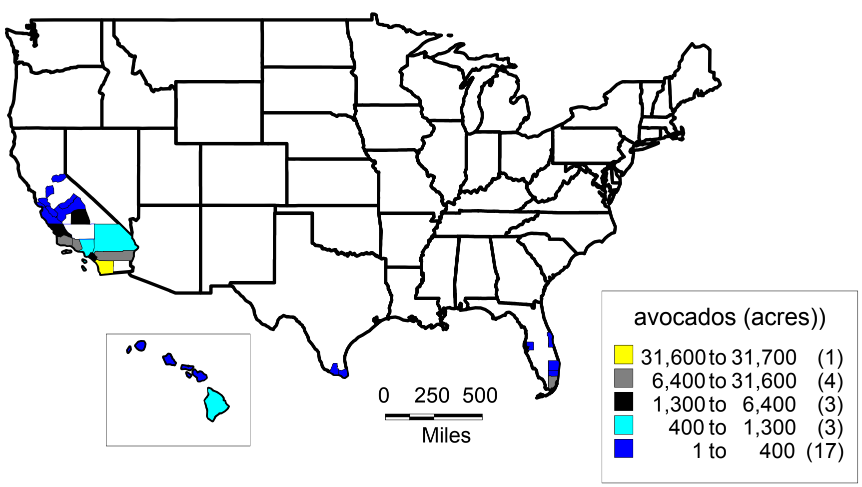


Figure 4c. Distribution of avocado orchard acreage in the United States (all commercial cultivars).

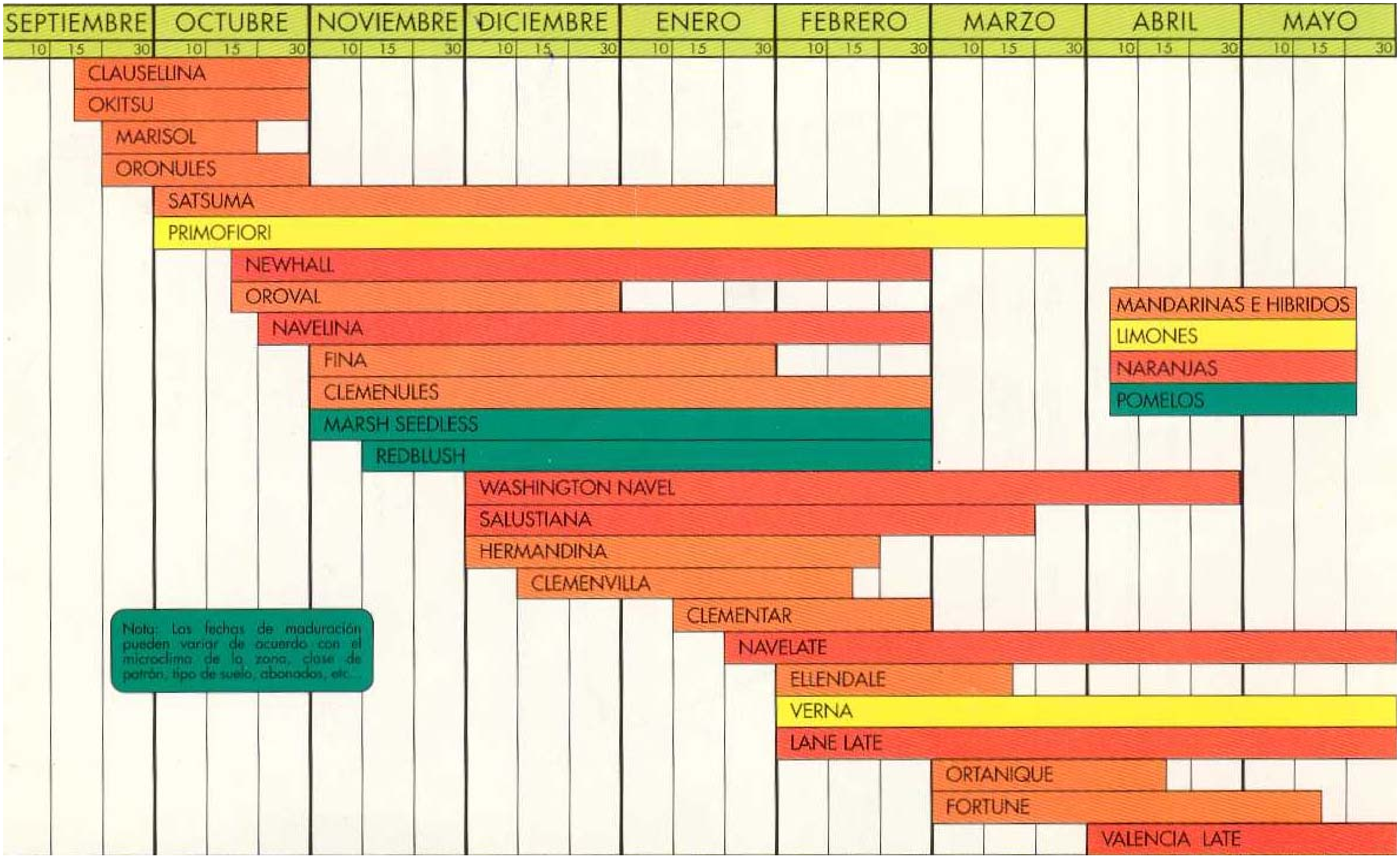


Figure A.2.1. Phenological timing of all citrus in Spain. Source: Santaballa, 2002.

XV. Appendices

Appendix 1*

The text presented here is an adaptation of the procedures recommended by FDA and described in www.fda.gov and www.foodsafety.gov.

Guidelines. The guidelines here represent a systematic approach to identifying, evaluating and controlling hazards. HACCP was developed in the area of food safety. However, its phases (principles) are broadly applicable and are used here with adaptations as appropriate to the area of quarantine safety or quarantine security.

A monitoring system as captured in PPQ's workplans is designed to emphasize prevention and control over reaction and remediation. Our intent is to implement the monitoring systems described here to prevent pest introduction and provide quarantine security. As in the area of food safety, USDA-APHIS-PPQ has been achieving this goal through a combination of regulatory and cooperative programs domestically and internationally. Whereas the purposes are indeed similar (prevention and reduction of the risk of a hazard), USDA-APHIS recognizes the value of the HACCP framework in assuring that key safeguarding elements are addressed. Hereafter, the guidelines are discussed as applicable to USDA-APHIS-PPQ. Elements that are redundant with existing USDA-APHIS procedures and guidelines (e.g., PRA guidelines) are not detailed.

Safeguarding (from pest introductions) is achieved by assessing the inherent hazards attributable to the importation of a commodity or other initiating action, determining the necessary steps that will control the identified hazards, and implementing active phytosanitary control practices to ensure that the hazards are eliminated or minimized.

Essentially, the workplan to be implemented by PPQ represent a system that identifies and monitors specific phytosanitary hazards – exotic pest species – that can adversely affect natural ecosystems and agricultural productivity. This hazard analysis serves as the basis for establishing critical control points (CCPs). CCPs identify those points in the process that must be controlled to ensure appropriate safeguards. Further, critical limits are established that document the appropriate parameters that must be met at each CCP. Monitoring and verification steps are included in the system, again, to ensure that potential hazards are controlled. The hazard analysis, critical control points, critical limits, and monitoring and verification steps are documented in a workplan. Seven principles have been developed which provide guidance on the development of an effective monitoring workplan.

(1) Acceptable level means the presence of a hazard that does not pose the likelihood of causing an unacceptable phytosanitary risk.

(2) Control point means any point in a specific pathway at which loss of control does not lead to an unacceptable phytosanitary risk.

(3) Critical control point, as defined here, means a point at which loss of control may result in an unacceptable phytosanitary risk.

(4) Critical limit, as defined here, means the maximum or minimum value to which a physical, biological, or chemical parameter must be controlled at a critical control point to minimize the risk that the identified phytosanitary hazard may occur.

(5) Deviation means failure to meet a required critical limit for a critical control point.

(6) Workplan, as defined here, means a written document that delineates the formal procedures for assuring phytosanitary safeguards and which is based on principles developed by The National Advisory Committee on Microbiological Criteria for Foods and modified for phytosanitary applications here.

(7) Hazard, as defined here, refers to an exotic pest that may cause an unacceptable phytosanitary risk.

(8) Monitoring means a planned sequence of observations or measurements of critical limits designed to produce an accurate record and intended to ensure that the critical limit maintains product safety. Continuous monitoring means an uninterrupted record of data.

(9) Preventive measure means an action to exclude, destroy, eliminate, or reduce a hazard and prevent recontamination through effective means.

(10) Risk means an estimate of the likely occurrence of a hazard.

(11) Verification means methods, procedures, and tests used to determine if the fielded production system and the shipping and distribution activities associated with the system are in compliance with the workplan.

A monitoring system as captured in the workplan will emphasize the industry's role in continuous problem solving and prevention rather than relying solely on periodic facility inspections by regulatory agencies.

The workplan offers two additional benefits over conventional inspection techniques. First, it clearly identifies importers and exporters as the final party responsible for ensuring the phytosanitary safety of commodities in trade. A workplan requires industry to analyze its production and pest management methods in a rational, scientific manner in order to identify critical control points and to establish critical limits and monitoring procedures. A vital aspect of industry's (or as represented by NPPOs) responsibility is to establish and maintain records that document adherence to the critical limits that relate to the identified critical control points, thus resulting in continuous self-inspection. Secondly, a workplan-based system allows the regulatory agency to more comprehensively determine industry's level of compliance. Use of a workplan in an import/export program requires development of a plan to address safeguards from pests. This plan must be shared with the regulatory agency because it must have access to CCP monitoring records and other data necessary to verify that the workplan is working. Using conventional inspection techniques, an agency can only determine conditions during the time of inspection, which provide a "snapshot" of conditions at the moment of the inspection. However, by adopting a dynamic, monitoring workplan approach, both current and past conditions can be determined. When regulatory agencies review workplan records, they have, in effect, a look back through time. Therefore, the regulatory agency can better ensure that processes are under control.

Traditional inspection is relatively resource-intensive and inefficient and is reactive rather than preventive compared to the workplan approach for ensuring phytosanitary safeguards. Regulatory agencies are challenged to find new approaches to safeguarding that enable them to become more focused and efficient and to minimize costs wherever possible. Thus, the advantages of transparent guidelines including regulatory inspections are becoming increasingly acknowledged by the regulatory community.

<u>HACCP background</u>. Established in 1988, the National Advisory Committee on Microbiological Criteria for Foods (NACMCF) is an advisory committee chartered under the U.S. Department of Agriculture (USDA) and comprised of participants from the USDA (Food Safety and Inspection Service), Department of Health and Human Services (U.S. Food and Drug Administration and the Centers for Disease Control and Prevention), the Department of Commerce (National Marine Fisheries Service), the Department of Defense (Office of the Army Surgeon General), academia, industry and state employees. NACMCF provides guidance and recommendations to the Secretary of Agriculture and the Secretary of Health and Human Services regarding the microbiological safety of foods.

(B) Development of HACCP Principles

In November 1992, NACMCF defined seven widely accepted HACCP principles that were to be considered when developing a HACCP plan. In 1997, the NACMCF reconvened the HCCP Working Group to review the Committee's November 1992 HACCP document and to compare it to current HACCP guidance prepared by

The CODEX Committee on Food Hygiene. From this committee, HACCP was defined as a systematic approach tot he identification, evaluation and control of food safety hazards based on the following seven principles:

Principle 1: Conduct a hazard analysis.
Principle 2: Determine the critical control points (CCPs).
Principle 3: Establish critical limits.
Principle 4: Establish monitoring procedures.
Principle 5: Establish corrective actions.
Principle 6: Establish verification procedures.
Principle 7: Establish record-keeping and documentation procedures.

Description of workplan stages (principles).

The workplan stages are consistent with the principles embodied by HACCP but constitute guidelines for monitoring the application of phytosanitary measures necessary to provide adequate quarantine security.

Principle 1
Flow Diagram. A flow diagram that delineates the steps in the production system and transportation pathway forms the foundation for applying the seven principles. The significant hazards associated with each step in the flow diagram should be listed along with preventative measures proposed to control the hazards. This tabulation will be used under Principle 2 to determine the CCPs. The flow diagram should be constructed by a workplan team that has knowledge and expertise on the commodity and associated pests, pest management, and the likely hazards. Each step in a process should be identified and observed to accurately construct the flow diagram.

Developing Preventive Measures. The preventive measures procedure identifies the steps in the process at which hazards can be controlled.

After identifying the hazards, industry and regulatory agencies must then consider what preventive measures, if any, can be applied for each hazard. Preventive measures are phytosanitary and other pest control tactics that can be used to control an identified phytosanitary hazard. More than one preventive measure may be required to control a specific hazard and more than one hazard may be controlled by a specified preventive measure.

Principle 2
Identify the critical control points (CCP) IN the pathway.

A CCP is a point, step, or procedure at which control can be applied and a phytosanitary hazard can be prevented, eliminated, or reduced to acceptable levels. Points in pathway that may be CCPs include hot treatment, cold treatment, fumigation, pest eradication, low prevalence, etc,

Principle 3
Establish critical limits for preventive measures associated with each identified CCP.

This step involves establishing a criterion that must be met for each preventive measure associated with a CCP. Critical limits can be thought of as boundaries of safety for each CCP and may be set for preventive measures such as temperature, time, pest densities, or number of bait sprays. Critical limits may be derived from sources such as regulatory standards and guidelines, scientific literature, experimental studies, and consultation with experts.

(a) Critical Limit

A critical limit is defined as a criterion that must be met for each preventive measure associated with a CCP. Each CCP will have one or more preventive measures that must be properly controlled to ensure prevention, elimination, or reduction of hazards to acceptable levels. Industry is responsible for using competent authorities to validate that the critical limits chosen will control the identified hazard.

(b) Target Level
In some cases, variables involved in the implementation of a phytosanitary measure may require certain target levels to ensure that critical limits are not exceeded. For example, a preventive measure and critical limit may be an internal fruit temperature of 2°C () during one stage of a process. The ship hold temperature, however, may be 2 ±2°C (); thus a ship hold target temperature would have to be less than -0°C () so that no product receives a cold treatment of more than 2°C ().

Principle 4.
Establish monitoring procedures.

Observations and Measurements
Monitoring is a planned sequence of observations or measurements to assess whether a CCP is under control and to produce an accurate record for use in future verification procedures. There are three main purposes for monitoring:

(i) It tracks the system's operation so that a trend toward a loss of control can be recognized and corrective action can be taken to bring the process back into control before a deviation occurs;
(ii) It indicates when loss of control and a deviation have actually occurred, and corrective action must be taken; and
iii) It provides written documentation for use in verification of the workplan.

Principle 5
Establish corrective actions.

(a) Purpose of Corrective Action Plan
Although the workplan-based system is intended to prevent deviations from occurring, perfection is rarely, if ever, achievable. Thus, there must be a corrective action plan in place to:
(i) Determine the disposition of any commodity that arrives at a port when a deviation occurred;
(ii) Correct the cause of the deviation and ensure that the critical control point is under control; and
(iii) Maintain records of corrective actions.

Principle 6

Establish procedures that verify that the workplan monitoring system is working.
(a) Establishing Verification Procedures

(i) The first phase of the process is the scientific or technical verification that critical limits at CCPs are satisfactory.
(ii) The second phase of verification ensures that the facility's workplan implementation plan is functioning effectively.
(iii) The third phase consists of documented periodic revalidations and modification, as necessary.
(iv) The fourth phase of verification deals with the regulatory agency's responsibility and actions to ensure that the establishment's workplan implementation system is functioning satisfactorily.

(b) The following are some examples of workplan verification activities:

(i) Verification procedures may include: Establishment of appropriate verification inspection schedules; Review of the work plan; Review of CCP records; Review of deviations and their resolution, including the disposition of commodities; Visual inspections of operations to observe if CCPs are under control; Random sample collection and analysis; Review of critical limits to verify that they are adequate to control hazards; Review of written record of verification inspections which certifies compliance with the workplan or deviations from the plan and the corrective actions taken; Validation of workplan, including on-site review and verification of flow diagrams and CCPs; and Review of modifications of the workplan.

(ii) Verification inspections should be conducted:

+ Routinely or on an unannounced basis, to ensure that selected CCPs are under control;
+ When it is determined that intensive coverage of a specific commodity is needed because of new information concerning new pests or new hazards associated with known pests; When treated commodities have been implicated as a means of entry of exotic pests;
+ When requested on a consultative basis and resources allow accommodating the request;
+ When established criteria have not been met; and
+ To verify that changes have been implemented correctly after a workplan has been modified.

(iii) Verification reports should include information about:
+ Existence of a workplan and the person(s) responsible for administering and updating the workplan; The status of records associated with CCP monitoring;
+ Direct monitoring data of the CCP while in operation; Certification that monitoring equipment is properly calibrated and in working order;
+ Deviations and corrective actions;
+ Any samples analyzed to verify that CCPs are under control. Analyses may involve physical, chemical, microbiological, or visual methods;
+ Modifications to the workplan; and
+ Training and knowledge of individuals responsible for monitoring CCPs.

(c) Training and Knowledge
(i) Focus and Objective
Training and knowledge are very important in making the workplan implementation successful in phytosanitary systems. Workplan-based systems work best when integrated into each employee's normal duties rather than added as something extra.
The depth and breadth of training will depend on the particular employee's responsibilities within the establishment. Management or supervisory individuals will need a deeper understanding of the workplan process because they are responsible for proper plan implementation and routine monitoring of CCPs such as cold treatment temperatures, pre-cooling, and treatment times. The training plan should be specific to the commodity being inspected rather than attempt to develop workplan expertise for broad application.
The inspector’s training should provide an overview of the workplan’s prevention philosophy while focusing on the specifics of the employee's normal functions. The CCPs such as proper equipment calibration and fruit inspection should be stressed. The use of Standard Operating Procedures (SOPs), which include the critical limits of treatments and treatment details, should be included.
For all employees, the fundamental training goal should be to make them proficient in the specific tasks that the workplan requires them to perform. This includes the development of a level of competency in their decision-making about the implementation of proper corrective actions when monitoring reveals violation of the critical limit. The training should also include the proper completion and maintenance of any records specified in the establishment's plan.

(ii) Reinforcement
Training reinforcement is also needed for continued motivation of the phytosanitary employees. Some examples might include:

+ A workplan video training program such as PPQ’s Safeguarding Video;
+ Changing reminders about workplan critical limits such as "No more than 2 degrees assures safe trade!" printed on employee's time cards or checks; and
+ Work station reminders such as pictorials on how and when to monitor temperatures or inspect fruit.

Every time there is a change in pest management or quarantine systems within the industry, the workplan training needs should be evaluated. The employees should be made sensitive to how the changes will affect phytosanitary safety

The workplan should include a feedback loop for employees to suggest what additional training is needed. All employees should be made a part of the continuous phytosanitary safety improvement cycle because the statement is very true: "The health of America's agriculture and natural systems is in their hands". This helps maintain their active awareness and involvement in the importance of each job to the safety of the traded commodities.

Principle 7
Establish effective record keeping systems that document the workplan

(a) Written workplan
This principle requires the preparation and maintenance of a written workplan by the regulatory organizations and industry. The plan must detail the hazards of each individual or categorical product covered by the plan. It must clearly identify the CCPs and critical limits for each CCP. CCP monitoring and record keeping procedures must be shown in the establishment's workplan. Workplan implementation strategy should be provided as a part of the producers/exporter's documentation.

(b) Record Keeping
The principle requires the maintenance of records generated during the operation of the plan. The record keeping associated with workplan procedures ultimately makes the system work. The requirement to record events at CCPs on a regular basis ensures that preventive monitoring is occurring in a systematic way. Unusual occurrences that are discovered as CCPs are monitored or that otherwise come to light must be corrected and recorded immediately with notation of the corrective action taken.

The level of sophistication of the record keeping necessary for the producers is dependent on the complexity of the production operation. Greenhouse operations will be in general more information intense than field operations.

(c) Contents of the Plan and Records
The approved workplan and associated records must be on file at the packinghouse or production area. Generally, the following are examples of documents that can be included in the total workplan based monitoring system:

(i) Listing of the workplan team and assigned responsibilities;
(ii) Description of the commodity and its intended distribution, destination and use;
(iii) Flow diagram for the pathway indicating CCPs;
(iv) Hazards associated with each CCP and preventive measures;
(v) Critical limits;
(vi) Monitoring system;
(vii) Corrective action plans for deviations from critical limits;
(viii) Record keeping procedures; and
(ix) Procedures for verification of workplan.
(d) Format for workplan information

*Disclaimer: USDA's extensive quoting from HACCP, identification of parallels, and incorporation of terminology that is more consistent with phytosanitary regulations is presented here as a means to illustrate how current methods used by USDA in its risk analyses and subsequent development of rules and regulatory workplans are consistent with standards used in other areas. USDA does not mean to imply incorporation of new regulations by reference to HACCP. Its presentation of the documentation in this appendix is intended to provide useful reference points and a framework that may help communicate the content, intent and sprit of USDA's regulatory workplans.

Appendix 2. Production of Clementines in Spain

[Document provided by MAPA's Dr. E. Santaballa]

1.- General information of Clementines MANDARINS cultivated economically in Spain

1.1.- Characteristics of Clementines mandarin varieties
The Clementines mandarin varieties highly cultivated in Spain are: Marisol, Oroval, Clemenules, Fina, Hernandina

The general characteristics of Clementines mandarins are
Medium sized, of a bright reddish colour and round or slightly flattened in shape. The skin is easily separated from the flesh, which is divided into about 11 large-celled sections. The Clementines is sweet and pungent and usually free from pips. It ripens from early November to mid-March.

The characteristics of different varieties of Clementines are
MARISOL
This most promising of Clementines selections originated as a bud mutation on Oroval in 1970 at Bechi in Castellon Province. Tree and fruit characteristics are indistinguishable from Oroval, with one significant exception: Marisol matures at least two weeks earlier than Oroval and is therefore as early as the Owari satsuma and seems destined to make inroads into these two varieties. This is already evident from its current popularity, with plantings of around 250.000 trees per year throughout Spain (or 15 per cent of all mandarins).
OROVAL
Oroval, a bud mutation of Fina, was found in 1950 at Quart de les Valls in Valencia Province, Spain. The trees are vigorous, well developed but thorny, although this characteristic declines with age. The fruit is only slightly larger than Nules and matures fully three weeks earlier. However, it has two important disadvantages from a production point of view: poor hanging ability because the rind, which has a somewhat more pebbly texture than Nules, becomes excessively puffy with delayed harvest; secondly, a rind which is susceptible to what is known locally as "water spot" following heavy rains, which causes the fruit to drop to the ground.

Although the flesh is reasonably tender and even more juicy than Nules, it is more acidic despite having good sugar levels. The urgency with which producers harvest the Oroval is sometimes reflected in poorer than optimum quality. This and other shortcomings have been noted by producers and are reflected in current plantings: only 1 per cent of all Clementines are of this variety. However, there are an estimated 7,000 ha in production at the present time.
CLEMENULES
The most popular Clementines selection in Spain where it constitutes around half of current plantings, Nules was discovered near the town of the same name in Castellón Province as a bud mutation on a Fina.

Like the Fina, Nules trees are vigorous, attain large size, and are very productive, out yielding the Fina by about 10 per cent. Moreover, the fruit is significantly larger (although somewhat smaller than the Oroval), maturing only a few days later than the Oroval), maturing only a few days later than Fina, in late November. An important characteristic of Nules is the extended period over which the fruit to be harvested until the end of January, if climatic conditions are favourable.

The extended harvesting period is made possible by up to three fruit sets, the fruit becoming more coarse and larger with each ser. Picking selectively is therefore an essential part of good management of Nules orchards. Packers and shippers will commonly pay a 15 to 20 per cent premium for Nules over Oroval, so much better is the quality.
FINA
First introduced into Spain in 1925, probably from Algeria, the Fina laid the foundations on which the country's Clementines industry developed. Until the early 1960s only Fina Clementines was grown on any scale in Spain. All other Spanish Clementines are derived from the Fina either directly or via one generation.

Fina trees are vigorous, dense and large and have good productivity. Although relatively later maturing by as much as four weeks compared with the early selections such as Marisol and Oroval, it is still the finest quality Spanish Clementine and is the one against which others are compared. Unfortunately the fruit is very small, much of the crop being below 60 mm in diameter (averaging 50 mm), with the result that market returns on a high proportion of smaller fruit cannot compete with other selections which produce larger is somewhat inferior fruit.

The rind is particularly smooth, and the fruit has excellent organoleptic characteristics: high juice content, very tender and sweet with good acid level but high sugar to acid ratio. It has the strong, pleasant aroma which typifies the Clementine.

Fruit may be left on the tree for relatively long period without noticeable quality deterioration. It is recommended for planting only in areas where soil and climate permit large size fruit. The Fina is no longer planted in Spain because of fruit size problems but around 10,000 ha are in production. Along with Nules, it is still the most extensively grown Clementine variety in Spain.

HERNANDINA

Discovered in 1966 as a bud mutation of Fina at Picasent in Valencia Province, the Hernandina is an exciting selection at present being extensively planted in the late areas of Spain.

Tree characteristics are almost the same as Fina, and so too are those of the fruits, with one important exception: the Hernandina's external colour develops two months later than the Fina. It is not harvested until mid-January and can be held in good condition and without quality deterioration until late February or early March.

Colour development is characteristically incomplete on a significant percentage of fruit with a small but acceptable area of the rind at the stylar-end remaining slightly green. Somewhat surprisingly the internal maturity is reached not more than one or two weeks later than the Fina and remains outstanding for an additional three months.

The Hernandina does not store well after harvest and may develop granulation if held on the tree past peak maturity. Nevertheless, price realisations on European markets have been most rewarding and have encouraged current planting rates of over 100,000 trees per year.

1.2.- Annual cultivating schedule of mandarin varieties and harvest time.

In Spain the cultivating schedule is very similar for all the varieties. The main cultural practices are:

Fertilization:

It is usually made in two times. The first one in March, and N, P, K and microelements are supplied. This one will be the only supply of P and K for the entire year. The amounts provided will depend on soils characteristics. The N will be provided in ammonia form. The quantity provided this time would be the 60 % of the whole year.

In the second supply only N, as N nitric, will be provided. Occasionally microelements can be provided, depending on trees.

The annual amounts of N are variable, but, as average, it can be provided 0,5 kg N / tree.

Pruning:

In Spain the pruning is made yearly, in March-April, when the risks of low temperatures have disappeared. The entire pruning in Spain is manual.

Irrigation:

The entire surface dedicated to mandarin cultivating is placed in irrigation areas.

The most commonly used method is trickle irrigation (70%), the rest (30%) by flood irrigation. When flood irrigation is used, 8 to 10 irrigations are given, starting in March - April an finishing in October - November.

Phytosanitary treatments:

They are detailed at point 6

Other cultural practices

They are usually started in March and finished in September

Harvest periods

Marisol: 15 Sep. – 15 Oct.
Oroval: 15 Oct. – 30 Dec.
Clemenules: 1 Nov. – 28 Feb
Fina: 1 Nov. – 30 Jan.
Hernandina: 1 Dec. – 28 Feb.

FIGURE A2. 1.- Maturation table of citrus fruits in Spain

1.3.- Major producing area of Clementine mandarin varieties and map.

The zones of higher production in Spain are located in the Comunidad Valenciana (provinces of Castellon, Valencia and Alicante) with 45.000 ha (87,5%) Murcia 2,2%), and Andalucia (provinces of Huelva, Sevilla and Cordoba) with 2300 ha.(4,5%) and Cataluña (Tarragona province) 3000 ha (5.8%).

The location of the production zones are represented in the figure 1

1.4.- Yield of each mandarine varieties.

The yield for trees at full production (10 years of plantation) oscillates in the 5 varieties among 25 and 30 tm/ha.

Figure 1.- Major Clementine producing areas in Spain

2.- Information of production of Clementine MANDARINS in the last several years.

The production (in tm) of Clementine mandarins in Spain in the last years has been the following, according to the data provided by Comite de Gestión de Frutos Cítricos.

Table 1.- Production of Clementine mandarins (in tm)

Variety	1996-97	1997-98	1998-99	1999-00	2000-01
Marisol	101.947	171.143	207.696	149.703	273454
Oroval	177829	195.465	181.769	174.254	142.730
Clemenules	447671	631.994	525.970	652.832	516.708
Fina	62240	67.573	61.140	74.493	55.262
Hernandina	95318	109.313	104.114	135.901	100.066
TOTAL	885.005	1.175.488	1.080.689	1.286.183	1.088.220

TABLE 2.-PRODUCTION OF MANDARINS IN THE DIFFERENT AREAS (IN TM) SEASON 2000/2001							
SPECIE/ VARIETY	Comunidad Valenciana	Región of Murcia	Comunidad Andaluza	Prov. de Tarragona	Baleares	Others	TOTAL
**MANDARIN							
*GROUP SATSUMAS	278.859	2.870	7.738	9.801	70	365	299.703
Clausellina-Okitsu	131.239	870	700		70		132.879
Satsuma	147.620	2.000	7.038	9.801		365	166.824
*GROUP CLEMENTINA	942.567	26.000	55.986	62.337	830	500	1.088.220
C. Marisol	259.554	13.900					273.454
C. Oroval	132.232	2.500	6.640	1.358			142.730
C. de Nules	444.831	6.000	19.100	46.447	830	500	516.708
C. Fina	26.624	2.100	12.006	14.532			55.262
C. Hernandina	80.326	1.500	18.240				100.066
*HYBRID MANDARIN	351.690	16.870	56.737	5.711	200	150	431.358
Clemenvilla Nova	126.342	3.440	4.599				134.381
Fortuna	123.063	11.000	17.815	1.164			153.042
Others	102.285	2.430	34.323	4.547	200	150	143.935
TOTAL MANDARIN	1.573.116	45.740	120.461	77.849	1.100	1.015	1.819.281

TABLE 3.-PRODUCTION OF MANDARINS IN THE DIFFERENT AREAS (IN TM) SEASON 1999/2000							
SPECIE/ VARIETY	Comunidad Valenciana	Región of Murcia	Comunidad Andaluza	Prov. de Tarragona	Baleares	Others	TOTAL
**MANDARIN							
*GROUP SATSUMAS	287.197	3.280	8.380	11.138	70	365	299.292
Clausellina-Okitsu	121.518	880	600		70		123.068
Satsuma	165.679	2.400	7.780			365	176.224
*GROUP CLEMENTINA	1.123.520	28.770	57.553	75.010	830	500	1.286.183
C. Marisol	233.833	15.570	300				149.703
C. Oroval	162266	2.300	8.258	1.430			174.254
C. de Nules	579.400	6.600	13.894	51.608	830	500	652.832
C. Fina	38170	2500	15.101	17.722			74.493
C. Hernandina	109.851	1.800	20.000	4.250			135.901
*HYBRID MANDARIN	378.115	18.690	58.481	1.200	200	150	456.836
Clemenvilla Nova	140.380	3.320	4.549				148.249
Fortuna	159.018	13.290	20.563	1.200			194.071
Others	78.717	2.080	33.369		200	150	114.516
TOTAL MANDARIN	1.788.832	50.740	124.414	87.348	1.100	1.015	2.042.311

TABLE 4.-PRODUCTION OF MANDARINS IN THE DIFFERENT AREAS (IN TM) SEASON 1998/1999							
SPECIE/ VARIETY	Comunidad Valenciana	Región of Murcia	Comunidad Andaluza	Prov. de Tarragona	Baleares	Others	TOTAL
**MANDARIN							
*GROUP SATSUMAS	262.364	3.980	6.155	7.605	70	365	280.539
Clausellina-Okitsu	116.253	980	200		70		117.503
Satsuma	146.111	3.000	5.955	7.605		365	163.036
*GROUP CLEMENTINA	975.588	34.960	35.819	32.992	830	500	1.080.689
C. Marisol	188.896	17.800	1.000				207.696
C. Oroval	168.914	3.500	8.371	984			181.769
C. de Nules	479.879	7.200	15.528	22.033	830	500	525.970
C. Fina	41.845	4.300	5.020	9.975			61.140
C. Hernandina	96.054	2.160	5.900				104.114
*HYBRID MANDARIN	349.707	16.370	21.706	4.852	200	150	392.985
Clemenvilla Nova	115.363	3.270	3.625				122.258
Fortuna	161.495	13.100	5.273	832			180.700
Others	72.849		12.808	4.020	200	150	90.027
TOTAL MANDARIN	1.587.659	55.310	63.680	45.449	1.100	1.015	1.754.213

TABLE 5.-PRODUCTION OF MANDARINS IN THE DIFFERENT AREAS (IN TM) SEASON 1997/1998							
SPECIES/ VARIETY	Comunidad Valenciana	Región of Murcia	Comunidad Andaluza	Prov. de Tarragona	Baleares	Others	TOTAL
**MANDARIN							
*GROUP SATSUMAS	293.515	5.074	7.218	6.041	50	200	312.098
Clausellina-Okitsu	87.739	1.200	1.230		50		90.219
Satsuma	205.776	3.874	5.988	6.041		200	221.879
*GROUP CLEMENTINA	1.078.069	24.319	31.928	39.125	1.750	300	1.175.491
C. Marisol	159.554	10.833	756				171.143
C. Oroval	182.739	2.400	9.301	1.028			195.465
C. de Nules	582.372	6.750	12.877	27.945	1.750	300	631.994
C. Fina	50.851	2.436	4.134	10.152			67.573
C. Hernandina	10.2553	1.900	4.860				109.313
*HYBRID MANDARIN	274.286	12.350	15.233	4.153	400	100	306.522
Clemenvilla Nova	108.556	2.800	3.625				114.981
Fortuna	121.211	9.300	5.525	691			136.727
Others	44.519	250	6.083	3.462	400	100	54.814
TOTAL MANDARIN	1.645.870	41.743	54.379	49.319	2.200	600	1.794.111

3.- Amount of Spanish Clementine MANDARINS for each usage and amount of export for each importing country for the last several years

3.1.- Usage of the Spanish Clementine mandarins

The distribution of this production, was the following (Table6):

Table 6.- Usage of the S Spanish Clementine mandarins (1000 tm)					
Season	Production	Exports	Domestic consumption		Withdrawal and wastes
			Fresh	Processing	
1996-97	885*	730,8	150	99	5,2
1997-98	1175*	895,9	261	156	88,7
1998-99	1080*	36,3	223	170	36,3
1999-00	1286,2	925,3	167,9	130	63
2000-01	1088	760	190	115	23
AVERAGE	1103*	669,66	198,38	134	43,24
%		56,19	16,64	11,24	3,63

*Author's (Santaballa) values corrected to correspond to Table 1.

3.2.- Importing country of Spanish citrus

The exports of Clementine mandarins per importing country is shown in the table 7

4.- The number of packing houses and producing groups.

In Spain there are around 600 citrus exporters. From these, around 500 (350 private exporters and 150 cooperative societies) export Clementine mandarins

However and because of technical and logistical complexities to export mandarines to U.S., it is estimated that only around 125 of these exporters have the capability to reach this objective

Table 7.- Importing countries of Spanish Clementine mandarins (In 1000 tm)					
COUNTRIES	SEASON				
	95-96	96-97	97-98	98-99	99-00*
FRENCH	205,0	194,2	211,4	162,1	195,1
GERMANY	265,0	239,8	287,4	229,1	260,2
NEDERLAND	34,3	42,4	53,5	40,5	47.6
BELGIUM	22,7	29,2	36,8	25,1	27.3
U. K. - IRELAND	52,2	53,9	62,6	50,1	66,4
DENMARK	8,8	10,8	16,3	13,2	15.5
SWEDEN	3,2	3,6	3,1	3,5	5.1
FINLAND	2,3	2,1	3,6	3,4	5.4
AUSTRIA	4,5	7,7	12,5	10,3	9.5
ITALY	3,5	25,3	29,0	33,4	47.5
PORTUGAL	0,8	1,0	2,4	5,6	1.8
TOTAL EEC	601,5	610,0	718,6	584,4	681.4
SWITZERLAND	24,4	25,2	28,8	25,5	18.3
NORWAY	4,0	3,8	9,2	9,9	9.2
TOTAL EUR. OC. OUSIDE EEC	28,4	29,0	38,0	35,4	27,5
USA	14,4	26,3	33,9	45,0	79,3
CANADA	7,1	8	8,2	7,5	8,9
ORIENTAL EUROPE	45,8	57,2	96,3	92,7	127,5
ANOTHER COUNTRIES	0,1	0,3	0,9	0,4	0,7
TOTAL OUTSIDE EU. 0C	66,8	91,8	139,3	619,8	216,4
TOTAL	696,7	730,8	895,9	765,4	925,3

5- Main MANDARINS pests: distribution.

The most important pests (11) and diseases (1) of the Clementine mandarins in Spain, and the periods of occurrence are shown below.

6.- Control method.

In the table 8 are shown the recommended products to treat the mentioned pests

Table 8.- Recommended products

Twospotted mite Tetranychus telarius	dicofol, dicofol+tetradifón, dicofol+exythiazox, fenbutatin, pyridaben, tebuphenpirad
Citrus red mite Panonychus citri	amitraz, dicofol+tetradifon, dicofol, exythiazox, fenbutatin, fenazaquin, flufenoxuron
Black scale Saissetia oleae	chlorfenvinphos, fenoxycarb, phosmet, methidathion, azinphos-methyl, piriproxyphen
Diaspine scales Parlatoria pergandei Lepidosaphes spp Aonidiella aurantii	Mineral oils, chlorpiriphos, azinphos-methyl, methidathion, omethoate, quinalphos, pirimiphos-methyl, piriproxyphen
Aphids A. ciotricola, A. Gossypii M. persicae, T. aurantii	Benfuracarb, carbosulfan chlorpiriphos dimethoate, ethiofencarb, metomyl, oxidemeton-methyl, pirimicarb,
Green bug Calocoris trivialis	dimethoate, malathion
Citrus leafminer Phyllocnistis citrella	abamectine, azadiractine benfuracarb, diflubenzuron, flufenoxuron hexaflumuron, imidaclorpid
Woolly Whitefly Aleurothrixus floccosus	buprofecin, butocarboxim,fenazaquin, fenotiocarb, flufenoxurón Mineral oils + ethion
Medfly Cerartitis capitata	Malathion
Phytophtora Root Rot	Copper compounds, Fosetyl-Al, Metalaxyl
Brown Rot Phytophthora spp	Copper compounds, Fosetyl-Al

*For the U.S., only the products in bold must be used

Appendix 3. Variability and Distribution of Input and Outputs [/1]

Part I. Model Output Characteristics

The table below details the simulation results after running 10,000 iterations of Monte Carlo sampling of the distributions described in tables 4a and 4b.

The outputs summarize the endpoints for the model. The values for the endpoints are described with the minimum, mean, and maximum values after 10,000 iterations. The inputs are similarly described.

The 95% confidence interval (last column) is interpreted as the value for which there is 95% confidence that values are equal to or below the number indicated. For example, in terms of the first row, the probability of a mated pair in a container under the mitigated scenario has a mean value of 3E-08 and further, 95% of the values associated with different iterations of this model will result in probability values equal to or less than 1.4E-07. The correspondence of this table with the results presented in Table 4d are indicated by marking the corresponding "result" numbers (1 and 2). The last two rows of the Output (labeled "Failures") correspond to Table 5c

Output Name	Minimum	Maximum	Mean	Std Dev	95%
Single Container-Mitigated, Prob. mated pair (Result 1, Mitigated)	4.0E-16	3.4E-06	3E-08	9.4E-08	1.4E-07
Single Container-Baseline Prob. mated pair (Result 1, Baseline)	6.1E-18	0.0002	2E-06	7.0E-06	9.3E-06
Multiple containers - Mitigated (Result 2, Mitigated)	1.2E-12	0.01	8.4E-05	0.0003	0.0004
Multiple containers- baseline (Result 2, Baseline)	0	0.4	0.006	0.02	0.03
Failures, P[Mated pair] single container	8.6E-14	0.02	0.0002	0.0007	0.001
Failures-P[Mated pair] multiple containers,	2.4354E-10	1	0.2	0.3	0.9

Input Name	Minimum	Maximum	Mean	Std Dev	95%
Mitigated / Number Fruit	108700	224978	166050	15373	191327
Mitigated / Fruit Infested with Larvae in the Field	1.3E-06	0.013	0.003	0.002	0.008
Mitigated / Larvae per fruit	1.0	7.7	3.5	1.3	5.8
Mitigated / Cold treatment	3.4E-09	9.0E-06	2.3E-06	1.6E-06	5.4E-06
Baseline / Number Fruit	108979	224260	166050	15372	191338
Baseline / Fruit Infested with Larvae in the Field	9E-07	0.13	0.02	0.02	0.07
Baseline / Larvae per fruit	1.0	7.7	3.5	1.3	5.8
Baseline / Cold treatment	2.5E-09	9.E-06	2.3E-06	1.6E-06	5.4E-06
Failures / Number Fruit	108025	223780	166050	15374	191327
Failures / Fruit Infested with Larvae in the Field	3.8E-06	0.13	0.03	0.02	0.07
Failures / Larvae per fruit	1.0	7.8	3.5	1.3	5.8
Failures / Cold treatment	2.2E-08	9.2E-05	2.3E-05	1.6E-05	5.4E-05

[/1]An MS Excel© spreadsheet with all parameters, inputs and calculations used for these simulations is available from the authors. The simulation was run using @Risk© software (Palisades, Inc.) but other software is also applicable. The MS Excel© program is sufficient to view the parameters and results.

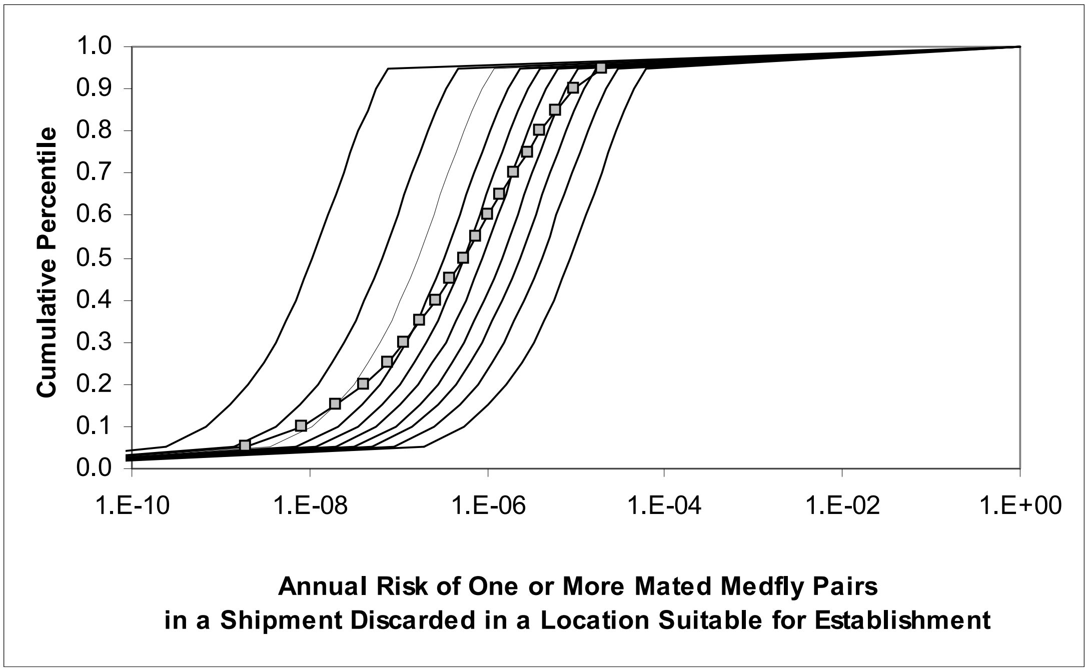
Part 2. Modeling Uncertainty and Variability.

Comments to early drafts of this document emphasized questions about separation of uncertainty and variability: conceptually, the difference between variability and uncertainty is clear. Variability refers to random variation that cannot be reduced through acquisition of additional information. Uncertainty refers to our state of knowledge; it may be reduced with additional information. A number of leaders in the field of risk analysis have drawn attention to cases where maintaining a rigorous distinction between uncertainty and variability, if possible, may be helpful in risk management decision-making. For example, if the statutory decisional criteria is "reasonable certainty of no harm," and this is administratively interpreted to mean protecting a hypothetical individual at the 99th percentile of the distribution of exposure to an environmental contaminant, then it may be necessary to consider the uncertainty associated with estimating this percentile in the exposure variability distribution. In this context, performing so-called 2-dimensional uncertainty analysis in which variable and uncertain model inputs are separated can lead to statements such as, "We are 95% confident that the individual at the 99th percentile in the exposure distribution does/not confront serious risk of illness." For evaluating the expected risk reduction potential of different phytosanitary strategies, however, it may be unnecessary to maintain a rigorous distinction between variability and uncertainty in risk analysis. Furthermore, while the conceptual and theoretical distinction between uncertainty and variability is clear, the separation can be somewhat artificial or vague in practice. Morgan (1998) cautions that while variability and uncertainty are different and sometimes require different treatments, the distinction can be overdrawn. In many contexts, variability is simply one of several sources of uncertainty (Morgan et al. 1990).

The National Research Council Committee that produced *Science and Judgment in Risk Assessment* (NRC 1994) acknowledged complications that arise because uncertainty and variability work in tandem: variability in one quantity can contribute to uncertainty in another, and the amount of variability is generally itself an uncertain parameter. Furthermore, this committee recognized that the lack of "identifiability" could frustrate efforts to partition variability and uncertainty. In the statistical sense, unidentifability means that the parameters of a model cannot be estimated from the available information. For example, a single observation consists of a variability component (how this individual varies from the population mean) and an uncertainty component (e.g., measurement error). If there are no matching replicates, a common problem in spatial or time series data, then it is impossible to empirically estimate the separate variability and uncertainty components. This problem has long been recognized, for example, in the field of geostatistics where it is referred to as the "nugget effect," where geological variation at a scale finer than the separation between measurement sites cannot be distinguished from uncertainty due to the survey protocol. Although various procedures have been developed in an effort to partition the "nugget" into variability and uncertainty, these procedures are themselves subject to uncertainty. Attempts to model "uncertainty about uncertainty" can lead to infinite regress. More recently, the National Research Council (NRC 2000) observed, "[a]lthough the distinction between natural variability and knowledge uncertainty is both convenient and important, it is at the same time hypothetical. The division of uncertainty into a component related to natural variability and a component related to knowledge uncertainty is attributable to the model developed by the analyst... Modeling assumptions may cause 'natural randomness' to become knowledge uncertainties, and vice versa."

Nevertheless, an effort was made to determine whether there was substantial informational value of a two-dimensional uncertainty analysis in the case of the risk management analysis for Spanish clementines. In this case, variability was assumed to dominate the total variation for all model inputs with the exception of the efficacy of cold treatment, where uncertainty was assumed to dominate. A two-dimensional (2-D) uncertainty analysis was conducted by extracting the 5th, 10th, ..., 90th, 95th percentiles of the cold treatment effect uncertainty distribution and running 19 separate Monte Carlo simulations of 5,000 iterations each. In the figure below, the results presented are for the annual risk of one or more mated *C. capitata* pairs being present in a shipment to a suitable geographic area and being discarded in a location suitable for potential establishment. The results of the 2-D uncertainty analysis are represented by a series of unmarked curves. These are presented in comparison to the result of a standard one-dimensional uncertainty analysis in which there is no distinction between uncertain and variable model inputs. The result of the 1-D uncertainty analysis is represented by the single curve marked with squares.

Comparison of 1-D v. 2-D Uncertainty Analysis



The outermost unmarked curves can be interpreted as delineating 90% confidence bounds (5^{th} – 95^{th} percent confidence levels) for each cumulative percentile of variability displayed on the y-axis. As shown in the figure, the 1-D analysis result is representative of the 2-D uncertainty analysis results. In the lower portion of the curve, the 1-D analysis is in the lower confidence region of the 2-D confidence bounds. In the upper portion of the curve, the 1-D analysis is in the upper confidence region of the 2-D confidence bounds. The 2-D analysis suggests that there is 95 percent confidence that the 95^{th} percentile of the variability distribution is less than approximately 6×10^{-5} (6.26 E-05). As indicated in the figure (and in the Summary Statistics table of Appendix 3 of the Summary Statistics Table above-part 1), the 95^{th} percentile of the 1-D analysis is approximately 2×10^{-5} (1.99 E-05). Given modeling results of the same order of magnitude (10^{-5}) and the presence of additional, unquantified uncertainties (e.g., the probability of establishment of a *C. capitata* colony, given one or more mated pairs discarded in a suitable location), the difference between the results is probably insubstantial and suggests that, at least in this case, the 2-D analysis provides little more than additional complexity.

Finally, additional evidence provided during the comment period (e.g., De Lima et al. 2002) has reduced the uncertainty about cold treatment efficacy and in this latest draft of the RMA document, most variability is considered attributable to measurable variability, as opposed to uncertainty.

N.B. Information in this section (Appendix 3, Part 2) incorporates comments and suggestions received from USDA's Office of Risk Assessment and Cost Benefit analysis (M. Powell, personal communication and USDA 2002).

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Appendix 4.

CLEMENTINES FROM SPAIN DECISION SHEET
USDA/APHIS/PPQ

Quarantine Pests That May Be Imported With Fresh Clementines (*Citrus reticulata*) Fruit From Spain:

The following insect pests are known to occur in Spain and are associated with clementines fruit. These US quarantine pests have been identified in this pest decision sheet as potential pests that may be imported with the commodity, clementines:

Ceratitis capitata (Wiedemann) (Diptera: Tephritidae)
Ceroplastes rusci (L.) (Homoptera: Coccidae)
Ceroplastes sinensis Del Guercio (Homoptera: Coccidae)
Cryptoblabes gnidiella (Milliere)(Lepidoptera: Pyralidae)
Parlatoria cinerea Hadden (Homoptera: Diaspididae)
Parlatoria ziziphi (Lucas) (Homoptera: Diaspididae)
Prays citri Milliere (Lepidoptera: Plutellidae)

SUMMARY:

Even though the seven quarantine pests listed above have the potential of being imported with clementines, all pests listed except *Ceratitis capitata*, would be easily detected by visual inspection during preclearance procedures. The scale insects, *Ceroplastes rusci, Ceroplastes sinensis, Parlatoria cinerea* and *Parlatoria ziziphi,* are relatively large and are located on the surface of the fruit. The larval stages of both Lepidopteran pests, *Cryptoblabes gnidiella* and *Prays citiri*, reside in or adjacent to the rind of the fruit. However, these two pests create large entrance holes in the fruit that are easily detected during a cursory inspection. This is not the case with the larvae of the Mediterranean fruit fly, *Ceratitis capitata.* They might not be detected during a visual inspection because the medfly larvae feed inside the fruit and the oviposition entrance holes are not readily visible. Supplement I provides a list of both arthropod and gastropod pests reported to be associated with citrus in Spain.

Of the twenty plant pathogens or the four parasitic nematode pests found, none are of quarantine significance. Twelve organisms that follow the pathway are non-quarantine pests, therefore no action is deemed necessary for the plant pathogens and pests listed in Supplement II.

Supplement I

Arthropod and Gastropod Pests Associated with *Citrus* sp. in Spain

Scientific Name	Geographic Distribution	Plant Part Affected	Quarantine Pest ?	Follow Pathway ?	Literature Cited/ Comments
ACARI					
Aceria sheldoni (Ewing) (Acari:Eriophyidae)	ES, CA, FL, HI	In	No	No	CABI, 2001
Brevipalpus obovatus Donnadieu (Acari:Tenuipalpidae)	ES, US	F, L, S	No	Yes	CABI, 2001; Jeppson *et al.*, 1975
Brevipalpus phoenicis (Geijskes) (Acari:Tenuipalpidae)	ES, FL	L, S	No	No	CABI, 2001
Panonychus citri (Acari: Tetranychidae)	ES,US	L,S	No	No	Baker and Tuttle, 1994
Panonychus ulmi Koch (Acari:Tetranychidae)	ES, US	L	No	No	CABI, 2001
Polyphagotarsonemus latus Banks (Acari:Tarsonemidae)	ES, US	F, Wp	No	Yes	CABI, 2001
Tetranychus cinnabarinus (Boisduval) (Acari:Tetranychidae)	ES, CA, TX	L	No	No	CABI, 2001
Tetranychus urticae Koch; (Acari:Tetranychidae) [synonym *T. telarius]*	ES, US	L	No	No	CABI, 2001
INSECTA					
Agrotis ipsilon (Hufnagel) (Lepidoptera:Noctuidae)	ES, US	Seedling	No	No	CABI, 2001
Agrotis segetum Denis and Schiffernuller (Lepidoptera:Noctuidae)	ES	Seedling	No	No	USDA, 1991
Aleurothrixus floccosus Maskell (Homoptera:Aleyrodidae)	ES, CA, TX, FL	F, In, L, S	No	Yes	CABI, 2001
Aonidiella aurantii (Maskell) (Homoptera:Diaspididae)	ES, CA, TX, AZ, FL	F, Wp	No	Yes	CABI, 2001
Apate monachus F. (Coleoptera:Bostricidae)	ES	S	Yes	No	CABI, 2001; USDA, 1991
Aphis craccivora Koch (Homoptera:Aphididae)	ES, US	B, In, L, Wp	No	Yes	CABI, 2001
Aphis fabae Scopoli (Homoptera:Aphididae)	ES, US	B, In, L, Wp	No	Yes	CABI, 2001
Aphis gossypii Glover (Homoptera:Aphididae)	ES, US	B, In, L, Wp	No	Yes	CABI, 2001
Aphis spiraecola Patch Synonym. *A. citricola* (Homoptera:Aphididae)	ES, US	F, In, Wp	No	Yes	CABI, 2001
Calocoris trivialis Costa	ES	S,L	Yes	No	Santaballa, 2002

Scientific Name	Geographic Distribution	Plant Part Affected	Quarantine Pest ?	Follow Pathway ?	Literature Cited/ Comments
(Hemiptera:Miridae)					(Appendix 2); Knight, 1968
Archips rosana (L.) (Lepidoptera:Tortricidae)	ES, OR	F, In, L	Yes	No	CABI, 2001;external fruit feeder
Aspidiotus nerii Bouche (Homoptera:Diaspididae)	ES, CA, HI	F, In, L	No	Yes	CABI, 2001
Asymmetrasca decedens (Paoli) (Homoptera:Cicadellidae)	ES	L	Yes	No	CABI, 2001; agrohispana. com, 2002
Brachycaudus helichrysi Kaltenbach (Homoptera:Aphididae)	ES, CA, ID	B, In, L	No	No	CABI, 2001; Bentley *et al.*, 2002
Cacoecimorpha pronubana (Hubner) (Lepidoptera:Tortricidae)	ES, OR	In, L	Yes	No	CABI, 2001; USDA, 1991
Ceratitis capitata (Wiedemann) (Diptera:Tephritidae)	ES	F	Yes	Yes	CABI, 2001; USDA, 1991
Ceroplastes floridensis Comstock (Homoptera:Coccidae)	ES, US	L, S	No	No	CABI, 2001
Ceroplastes rusci (L.) (Homoptera:Coccidae)	ES, FL	F, In, L, S, Wp	Yes	Yes	CABI, 2001; USDA, 1991
Ceroplastes sinensis Del Guercia (Homoptera:Coccidae)	ES, US	F, In, L, S, Wp	Yes	Yes	Ben-Dov, 2002; USDA, 1991
Charaxes jasius (L.) (Lepidoptera:Nymphalidae)	ES	L	Yes	No	CABI, 2001; Mazzei *et al.*, 2002
Chrysomphalus aonidum (L.) (Homoptera:Diaspididae)	ES, US	F, L, S	No	Yes	CABI, 2001
Chrysomphalus dictyospermi (Morgan) (Homoptera:Diaspididae)	ES, US	F, L, S	No	Yes	CABI, 2001
Coccus hesperidum (L.) (Homoptera: Coccidae)	ES, US	L, S	No	No	CABI, 2001
Cryptoblabes gnidiella (Milliere) (Lepidoptera:Pralidae)	ES	F	Yes	Yes	CABI, 2001; USDA, 1991
Dialeurodes citri (Ashmead) (Homoptera:Aleyrodidae)	ES, US	F, In, L, S	No	Yes	CABI, 2001
Diaspidiotus perniciosus (Comstock) (Homoptera:Diaspididae)	ES, US	F, Wp	No	Yes	CABI, 2001
Drosophila immigrans Sturtevant (Diptera:Drosophilidae)	ES, CA	Rotting fruit	No	No	CABI, 2001
Drosophila simulans Sturtevant (Diptera:Drosophilidae)	ES, US	Rotting fruit	No	No	CABI, 2001

Scientific Name	Geographic Distribution	Plant Part Affected	Quarantine Pest ?	Follow Pathway ?	Literature Cited/ Comments
Emposaca decipens Paoli (Homoptera:Cicadellidae)	ES	L	Yes	No	USDA, 1991
Emposaca vitis (Gothe) (Homoptera:Cicadellidae)	ES	L	Yes	No	USDA, 1991
Frankliniella occidentalis (Pergande) (Thysanoptera:Thripidae)	ES, US	In, L	No	No	CABI, 2001
Helicoverpa armigera (Hubner) (Lepidoptera:Noctuidae	ES	B, F, In, L	Yes	No	CABI, 2001; USDA, 1991 immature fruit
Heliothrips haemorrhoidalis Bouche (Thysanoptera:Thripidae)	ES, US	F, L	No	Yes	CABI, 2001
Hemiberlesia lataniae (Signoret) (Homoptera:Diaspididae)	ES, US	F, L, S	No	Yes	CABI, 2001
Icerya purchasi Maskell (Homoptera:Margarodidae)	ES, US	In, Wp	No	Yes	CABI, 2001
Lepidosaphes beckii (Newman) (Homoptera:Diaspididae)	ES, US	F, Wp	No	Yes	CABI, 2001
Lepidosaphes gloverii (Packard) (Homoptera:Diaspididae)	ES, US	F, L, S	No	Yes	CABI, 2001
Limothrips cerealium (Haliday) (Thysanoptera:Thripidae)	ES, US	Seeds	No	No	CABI, 2001
Aleurothrixus floccosus (Maskell) (Homoptera: Aleyrodidae)	ES, US	L,S,	No	No	CIE. 1974. Map 327
Macrosiphum euphorbiae (Thomas) (Homoptera:Aphididae)	ES, US	Wp	No	Yes	CABI, 2001
Myzus persicae Sulzer (Homoptera:Aphididae)	ES, US	In, Wp	No	Yes	CABI, 2001
Neoaliturus haematoceps (Mulsant and Rey) (Hemiptera:Cicadellidae)	ES	L	Yes	No	CABI, 2001
Neoaliturus tenellus (Baker) (Hemiptera:Cicadellidae)	ES, US	L	No	No	CABI, 2001
Nezara viridula (L.) (Hemiptera:Pentatomidae)	ES, US	F, Seeds, Wp	No	No	CABI, 2001
Nipaecoccus nipae (Maskell) (Homoptera:Pseudococcidae)	ES, CA, FL, LA	F, In, L, S	No	Yes	CABI, 2001
Orthezia insignis Browne (Homoptera:Ortheziidae)	ES, US	In, Whp	No	Yes	CABI, 2001
Pantomorus cervinus (Boheman) (Coleoptera:Curculionidae)	ES, US	L, R	No	No	CABI, 2001
Parabemisia myricae (Kuwana) (Homoptera:Aleyrodidae)	ES, CA, FL	L, S	Yes	No	CABI, 2001; Hamon *et al*., 2002
Parasaissetia nigra (Nietner)	ES, US	L, S	No	No	CABI, 2001

Scientific Name	Geographic Distribution	Plant Part Affected	Quarantine Pest ?	Follow Pathway ?	Literature Cited/ Comments
(Homoptera:Coccidae)					
Aonidiella aurantii(Maskell) (Homoptera:Diaspididae)	ES,US	L,S	No	No	CIE, 1968, Map 2
Parlatoria pergandii Comstock (Homoptera:Diaspididae)	ES, US	F, In, Wp	No	Yes	CABI, 2001
Parlatoria cinerea Hadden (Homoptera:Diaspididae)	ES	F	Yes	Yes	McKenzie, 1945; USDA, 1991
Parlatoria ziziphi (Lucas) (Homoptera:Diaspididae)	ES	F, L, S	Yes	Yes	CABI, 2001; McKenzie, 1945; USDA, 1991
Parthenolecanium corni (Bouche) (Homoptera:Coccidae)	ES, US	In, Wp	No	Yes	CABI, 2001
Peridroma saucia (Hubner) (Lepidoptera:Noctuidae)	ES, US	F, Seeds, Wp	No	Yes	CABI, 2001
Phyllocnistis citrella Stainton (Lepidoptera:Gracillariidae)	ES, AL, FL, LA, TX	L	Yes	No	CABI, 2001
Planococcus citri (Risso) (Homoptera:Pseudococcidae	ES, US	F, In, R, Wp	No	Yes	CABI, 2001
Prays citri Milliere (Lepidoptera:Plutellidae)	ES	F, In, L	Yes	Yes	CABI, 2001; USDA, 1991
Pseudococcus calceolariae (Maskell) (Homoptera:Pseudococcidae)	ES, CA, LA	F, In L, R	No	Yes	CABI, 2001
Pseudococcus longispinus (Targinoi-Tozzetti) (Homoptera:Pseudococcidae)	ES, US	F, In, L, S	No	Yes	CABI, 2001
Aphis gossypii (Homoptera:Aphididae)	ES,US	L,S	No	No	CIE, 1968, Map 18
Toxoptera aurantii. (Homoptera:Aphididae)	ES,US	L,S	No	No	CIE, 1961, Map 131
Rhopalosiphum maidis (Fitch) (Homoptera:Aphididae)	ES, US	In, L, S	No	No	CABI, 2001
Saissetia coffeae (Walker) (Homoptera:Coccidae)	ES, US	L, S	No	No	CABI, 2001
Saissetia oleae (Olivier) (Homoptera:Coccidae)	ES, US	L, S	No	No	CABI, 2001
Schistocerca gregaria (Forskal) (Orthoptera:Acrididae)	ES	Seeds, Wp	Yes	No	CABI, 2001; USDA, 1991 external feeder
Spodoptera exigua (Hubner) (Lepidoptera:Noctuidae)	ES, US	F, In, L	No	No	CABI, 2001; external feeder
Spodoptera littoralis (Boisduvall) (Lepidoptera:Noctuidae)	ES	F, L,	Yes	No	CABI, 2001; USDA, 1991

Scientific Name	**Geographic Distribution**	**Plant Part Affected**	**Quarantine Pest ?**	**Follow Pathway ?**	**Literature Cited/ Comments**
					external feeder
Sphrageidus similis Fuessly (Lepidoptera:Lymantriidae)	ES	L	Yes	No	USDA, 1991
Thrips flavus Schrank (Thysanoptera:Thripidae)	ES	In, Seeds, Wp	Yes	No	CABI, 2001
Toxoptera aurantii Boyer de Fonscolombe (Homoptera:Aphididae)	ES, US	B, In, L	No	No	CABI, 2001
Trichoplusia ni (Hubner) (Lepidoptera:Noctuidae)	ES, US	L, Wp	No	No	CABI, 2001
Xestia c-nigrum (L.) (Lepidoptera:Noctuidae)	ES, US	L, Seeds, Wp	No	No	CABI, 2001
GASTROPODA					
Theba pisana (Muller) (Pulmonata:Helicidae)	ES	Wp	Yes	No	CABI, 2001

Geographic Distribution: ES=Spain, US=United States. Individual states in US: AL=Alabama, AZ=Arizona, CA=California, FL=Florida, HI=Hawaii, ID=Idaho, LA=Louisiana, OR=Oregon and TX=Texas.

Plant Part Affected: B=Buds, F=Fruit, In=Inflorescence, L=Leaves, R=Roots, S=Stems, Wp=Whole Plant.

Highlighted rows indicate pests that will follow the pathway and are quarantine pests.

Supplement II

Pests Associated With Commodity in Country
Pathogens Associated With Citrus in Spain

Organism	**Geographic Distribution**[1]	**Plant Part Affected**[2]	**Quarantine Pest**[3]	**Follow Pathway**[3]	**References/ Comments**
BACTERIA					
Rhizobium radiobacter (Beijerinck & van Delden) Pribram*.* = *Agrobacterium tumefaciens* (Smith and Townsend) Conn (Proteobacteria: Rhizobiales)	ES, US	Wp	N	N	CABI, 2001
Pseudomonas syringae pv. *syringae* van Hall (Proteobacteria: Pseudomonadales)	ES, US	F, L, T, R	N	Y	CABI, 2001; APS, 2000, Compendium of Citrus Diseases, 2nd Ed.
FUNGI					
Alternaria brassicae (Berk.) Sacc. (Fungi Imperfecti: Hyphomycetes)	ES, US	F, L	N	Y	CABI, 2001
Alternaria citri Ellis & N. Pierce in N. Pierce (Fungi Imperfecti: Hyphomycetes)	ES, US	F, L, R, Tw	N	Y	CABI, 2001; APS, 2000, Compendium of Citrus Diseases, 2nd Ed
Aspergillus niger v. Tiegh (Fungi Imperfecti: Hyphomycetes)	ES, US	F, L	N	Y	CABI, 2001; APS, 2000, Compendium of Citrus Diseases, 2nd Ed
Botrytis cinerea Pers. ex Fr. [telemorph= *Botryotinia fuckeliana (de Bary) Whetzel]* *[=Sclerotinia fuckeliana* (de Bary) Fuckel*]*	ES, US	F, L, In, Tw	N	Y	CABI, 2001; APS, 2000, Compendium of Citrus Diseases, 2nd Ed
Diaporthe citri F.A. Wolf = *Phomopsis citri* H. Fawc. (Ascomycota: Diaporthales) Melanose	ES, US	F, L, Tw	N	Y	Arpaia & Kader, 1999; CABI, 2001; EPPO, 2001; APS, 2000, Compendium of Citrus Diseases, 2nd Ed
Giberella fujikori (Sawada) S. Ito (Ascomycota: Hypocreales) [syn=*Fusarium moniliforme* (J. Sheld)]	ES, US	F	N	Y	CABI, 2001
Lasiodiplodia theobromae (Pat.) Griffon & Maubl.	ES, US	F, T, R, Tw	N	Y	CABI, 2001; APS, 2000, Compendium

Organism	**Geographic Distribution**[1]	**Plant Part Affected**[2]	**Quarantine Pest**[3]	**Follow Pathway**[3]	**References/ Comments**
(anamorph *Botryodiplodia theobromae* Pat. and *Diplodia natalensis* Pole-Evans; teleomorph *Botrysphaeria rhodina* (Cooke) (Arx) (Fungi Imperfecti: Coelomycetes)					of Citrus Diseases, 2nd Ed
Ascomycota: Hypocreales) [anamorph=*Fusarium solani* (Mart.) Sacc.]	ES,US	F,Tw,St	N	N	CABI, 2001; APS, 2000, Compendium of Citrus Diseases, 2nd Ed
Penicillium italicum Wehmer (Fungi Imperfecti: Hyphomycetes)	ES, US	F	N	Y	CABI, 2001; APS, 2000, Compendium of Citrus Diseases, 2nd Ed
Phytophthora cactorum (Lebert & Cohn) Schroter (Oomycota: Pythiales)	ES, US	F, L, T, R	N	Y	CABI, 2001; APS, 2000, Compendium of Citrus Diseases, 2nd Ed
Phytophthora citrophthora (R.E.Sm.& E.H. Sm.) Leonian	ES, US	F, L, T, R	N	Y	CABI, 2001; APS, 2000, Compendium of Citrus Diseases, 2nd Ed
Phytophthora nicotianae Breda de Haan (Oomycota: Pythiales)	ES, US	F, L, T, R	N	Y	CABI, 2001; APS, 2000, Compendium of Citrus Diseases, 2nd Ed
Rosellinia necatrix Prill. (anamorph=*Dematophora necatrix* R. Hartig) (Ascomycota: Xylariales)	ES, US	R	N	N	CABI, 2001
VIRUS					
Apple stem grooving virus (ASGV) [Syn. =Citrus tatter leaf virus] (Capillovirus)	ES, US	Wp	N	N	CABI, 2001; U of Fl, CES, 1993, Citrus & Citrus Like Diseases, Bul. 18p.
Citrus exocortis viroid (CEVd) (Pospiviroidae: Pospiviroid)	ES, US	Wp	N	N	CABI, 2001; U of Fl, CES, 1993, Citrus & Citrus Like Diseases, Bul. 18p; APS, 2000, Compendium of Citrus Diseases, 2nd Ed
Citrus psorosis Complex	ES,US	Wp	Y	N	CABI, 2001; U of

Organism	Geographic Distribution[1]	Plant Part Affected[2]	Quarantine Pest[3]	Follow Pathway[3]	References/ Comments
Citrus psorosis virus A (CPsV-A) concave gum-blind pocket virus *Citrus psorosis virus B* (CPsV-B) (Syn.=Citrus ringspot disease) (Ophiovirus: Ophiovirus)					Fl, CES, 1993, Citrus & Citrus Like Diseases, Bul. 18p; APS, 2000, Compendium of Citrus Diseases, 2nd Ed
Citrus triseteza virus (CTV) (Closteroviridae: Closterovirus)	ES,US	Wp	N	N	CABI, 2001; U of Fl, CES, 1993, Citrus & Citrus Like Diseases, Bul. 18p; APS, 2000, Compendium of Citrus Diseases, 2nd Ed
Citrus viroid II (CVd-II) [Syn=*Citrus cachexia viroid*] (Pospiviroidae: Hostuviroid)	ES, US	Wp	N	N	CABI, 2001; U of Fl, CES, 1993, Citrus & Citrus Like Diseases, Bul. 18p.
NEMATODA					
Helicotylenchus dihystera (Cobb) Sher (Tylenchida: Hoplolaimidae)	ES, US	R	N	N	CABI, 2001
Tylenchulus semipenetrans Cobb (Tylenchida: Tylenchulidae)	ES, US	R	N	N	CABI, 2001
Xiphinema americanum Cobb. (Dorylaimida: Longidoridae)	ES, US	R	N	N	CABI, 2001
Xiphinema index Thorne & Allen (Dorylaimida: Longidoridae)	ES, US	R	N	N	CABI, 2001

Table Footnotes

[1]Only the distribution of pests in both Spain and the United States is considered. Geographic distribution legend: CA=California, ES=Spain, FL=Florida, ID=Idaho, LA=Louisiana, TX=Texas, US=United States. (ES) = Requires input from Spain.
[2]Plant part affected legend: Bk=Bark, Br=Branch, F=Fruit, In=Inflorescence, L=Leaf, R=Root, Sh=Shoot, St=Stem, T=Trunk, Tw=Twig, Wp=Whole plant
[3] Y=Yes, N=No

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